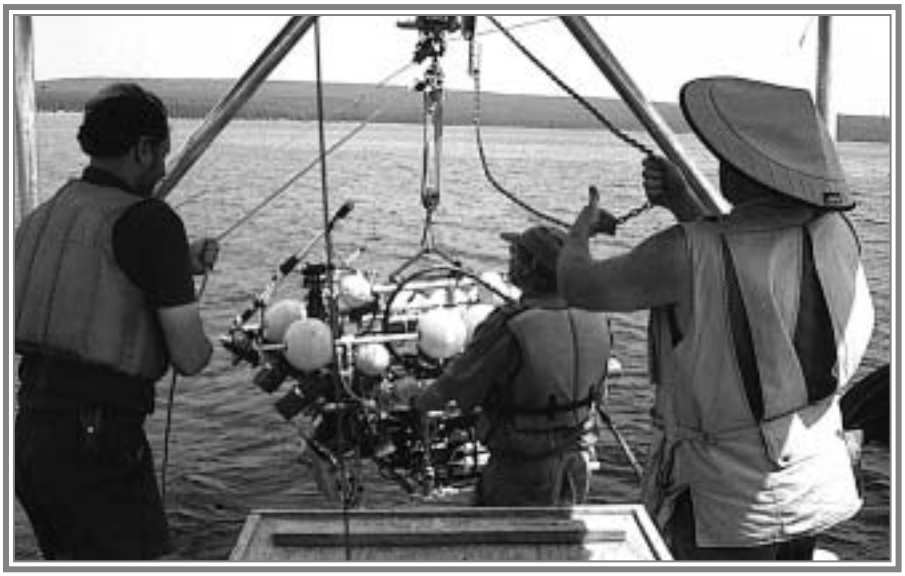
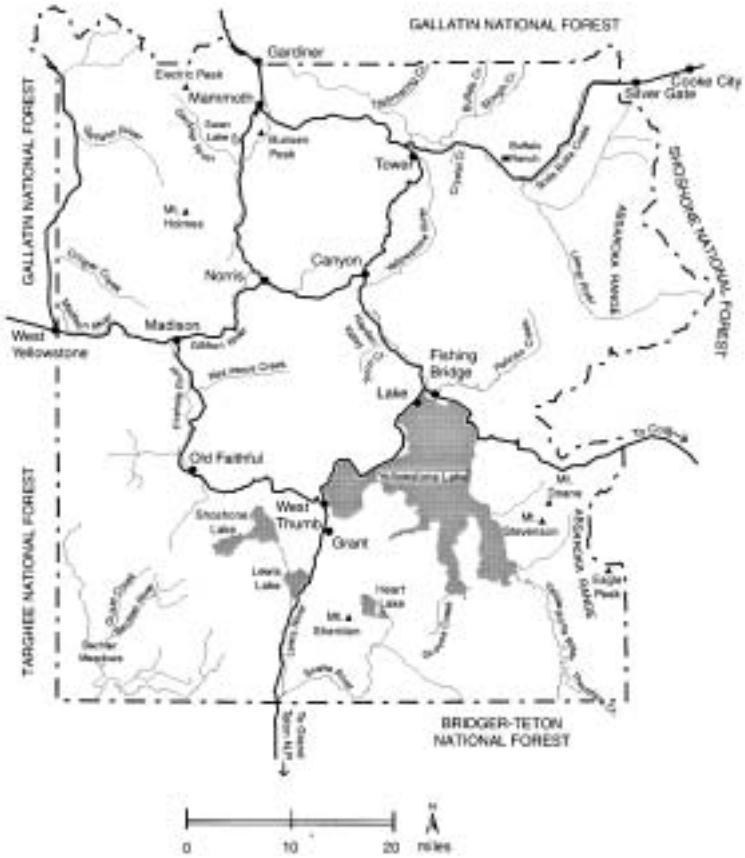
INVESTIGATORS' ANNUAL REPORTS 2000



YELLOWSTONE NATIONAL PARK



YELLOWSTONE NATIONAL PARK

Yellowstone Center for Resources
P.O. Box 168
Yellowstone National Park, Wyoming 82190

December 2002

YCR Annual Report: YCR-IAR-2002

Cover: USGS geoscientists launch a remotely-operated-vehicle as part of their investigations of hydrothermal features on the bottom of Yellowstone Lake. NPS photo.

Acknowledgements: The National Park Service thanks the researchers that have contributed to our knowledge and understanding of Yellowstone. This report was compiled and edited by Christie Hendrix and Alice K. Wondrak.

FOREWORD

Since the dawn of scientific wondering, human inquiry has led to the exploration, and often alteration, of almost everything in our world, at every scale—from the landscape of the moon to the human genome. In the national parks, however, through varying definitions and to varying degrees of success, we have attempted to "preserve natural conditions" for the past 130 years.

Their long-term preservation of natural resources makes parks reservoirs of information of great value to humanity, and perhaps today more than ever before, America's national parks are being recognized as being more than pleasuring grounds and nature preserves. The NPS's Natural Resource Challenge urges that in addition to using science as a means to improve park management, parks can and should be centers for broad scientific research and inquiry.

The national parks have long-captured the imagination of scientists, who recognized them as places where we could observe natural processes operating in places that had been less subject to human alteration than most others throughout the nation, and indeed throughout the world. In Yellowstone, those kinds of observed processes have ranged from macro-scale studies of landscape changes affecting the local ecosystem to micro-scale studies of tiny organisms that have the potential to change the lives of people the world over, making the protection of this wilderness relevant and crucial even to those who will never know its aesthetic and recreational wonders.

There are more than 300 index entries in this year's Investigators' Annual Report. That is a lot of science; a lot of knowledge being collected that needs to be shared. This report should not be seen as the body of that knowledge, but rather as its skeleton. Contact information is provided so that readers may learn more about the projects and results described here. All persons who wish to conduct their own research in Yellowstone are required to apply for a permit. Information on permitting procedures is available from the Research Permitting Office, Yellowstone Center for Resources, P.O. Box 168, Yellowstone National Park, WY 82190.

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ANTHROPOLOGY

Project title: People and Nature: Yellowstone as Landscape

Principal investigator: Mr. Raymond Fenio
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Unionville, IN 47468

Objective: This research will provide an ethnographic study of a national park. The descriptive data provided by ethnography are not easily assembled by other methods. By living in a place and observing, as well as talking with the people who visit, work, and reside within it provides a greater depth of understanding. In discovering what people do when they experience Yellowstone, what they expect and how these expectations shape their attitudes about parks, wilderness and recreation, this research should provide a valuable contribution to park management and hopefully encourage and invite thoughtful dialogue over what culture and nature means and their relative value in emerging policy.

Findings: This research is ongoing. However, some interviews were conducted with visitors and employees. Observations about what visitors actually do have been recorded but without any results or conclusions at this time.

ARCHEOLOGY

Project title: Geochemical Investigations of Obsidian Source Material

Principal investigator: Mr. Kenneth Cannon
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100 Centennial Mall North
Lincoln, NE 68508-3873

Additional investigator(s): Richard Hughes

Objective: To collect provenence and geochemical data on geologic sources of toolstone quality obsidian. This database will be used to compare geochemical data of artifacts for discerning aboriginal use of obsidian sources. This information will be useful in determining patterns of lithic procurement and land use in the Greater Yellowstone Ecosystem and beyond.

Findings: No fieldwork was conducted in YNP during 2000.

Project title: Archeological Research in Yellowstone National Park

Principal investigator: Dr. Leslie Davis
Phone number: 406-994-6614
Address: Museum of the Rockies
Montana State University
Bozeman, MT 59717-0272

Additional investigator(s): Brian Reeves

Objective: To carry out cultural resource compliance National Register testing, testing for preparation of data recovery plans, inventory in support of the trails rehabilitation program, salvage of eroding sites 48YE252 and 48YE409, and shoreline inventory on Yellowstone Lake.

Findings: Sites on the highway between Mammoth and Gardiner were evaluated for their significance. Some sites in this corridor remain to have their subsurface remains evaluated.

Trails work included inventory around Heart Lake, Jones Pass, and Warm Creek-Pebble Creek. Campground Site 24YE26 in the Black Canyon of the Yellowstone was tested and found to contain multiple stratified (n=6) components with intact features. The Pelican Lake (Late Archaic Period) occupations dominate the assemblages with earlier and later (possibly McKean) camps present. Of particular interest was the recovery of 3 large corner notched (hafted) knives manufactured from local (Hellroaring drainage)

chert/chalcedony.

Site 48YE252 was found to be multicomponent and being severely damaged by wave action on Yellowstone Lake. Judging from the artifact collection made on the beach, the site was occupied from Lake Paleoindian through Late Prehistoric times (~ 8000 BC to ~ AD 1500). Pelican Lake artifacts predominate. The hearth was radiocarbon dated to Pelican Lake culture (1970+/-60 BP).

Another eroding site, 48YE409, called Osprey Beach, was assigned to the Cody Complex and radiocarbon dated. Surface collecting and testing identified tools including 3 Cody Knives, a Scottsbluff point, and a sandstone shaft straightener. Examination of the geological profile by Drs. Ken Pierce (USGS) and Paul Doss (Yellowstone National Park geologist) suggests the Cody Complex people were camping on the Yellowstone Lake beach.

Data are accumulating for an obsidian sourcing project. This year, there is interesting information from the area south of Yellowstone Lake and the Yellowstone Lake shore. The southern portion of the park shows use by people using more Teton Pass and other southern obsidian sources, while the northern portion is dominated by Obsidian Cliff with Bear Gulch (Idaho) at about 7 percent. Specimens from Brown's Bench (southcentral Idaho on the Nevada border) and Crescent H (south of Wilson, Wyoming) were identified in Yellowstone archeological specimens for the first time this year.

Project title: FY00 Federal Highway Archeological Investigation in Yellowstone National Park

Principal investigator: Mr. David Eckles
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Additional investigator(s): Paul H. Sanders

Objective: Archeological inventory along the north and south rim roads that border the Grand Canyon of the Yellowstone at Canyon; archeological test excavation of eight prehistoric sites along the Canyon to Lake Junction road; and additional site recording at historic sites 48YE23 (Canyon Incinerator) and 48YE155 (Canyon Transportation Complex).

Findings: The archeological inventory resulted in the recording of a historic cistern, two historic trash dumps, and remnants of the Canyon Lodge. Except for a small portion of a buried trash level dating to the Shaw and Powell occupation at the Canyon Lodge, all other properties are recommended as not eligible for nomination to the National Register of Historic Places.

The test excavations of the eight prehistoric sites found that they represent lithic scatters of varying sizes and date from the Early Archaic to the Late Prehistoric periods, primarily based on diagnostic artifacts. One hearth feature was uncovered at 48YE545 which dated at 1070 years BP. Except for the latter site, few clear buried cultural levels were encountered.

The site investigation of historic sites 48YE23 and 48YE155 resulted in the recording of a number of

features and a sparse amount of historic debris. Both sites had been razed, with few intact historic deposits. Both sites are recommended as not eligible due to the lack of integrity.

Project title: Chemical Analysis of Obsidian Sources and Artifacts from the Northwest and Great Plains, USA

Principal investigator: Dr. Michael D. Glascock
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Address: Research Reactor Center
University of Missouri
Columbia, MO 65211
Additional investigator(s): Craig E. Skinner

Objective: The major objective of this research is to establish a geochemical database of obsidian sources in the northwestern USA, including sources in Yellowstone National Park and adjoining areas. Samples of obsidian from various source areas were collected and analyzed by three techniques (neutron activation analysis and x-ray fluorescence analysis and inductively coupled plasma-mass spectrometry) to establish the database. The geographic coordinates of each sample have been entered into the database along with the chemical analysis information. Artifacts from locations great distances from Yellowstone are now being analyzed similarly and compared to the source database. Patterns of obsidian exchange are being established.

Findings: In fall 2000 and continuing through 2001 we are utilizing a new and less destructive analytical technique to study obsidian source and artifact materials. This new method is laser ablation-inductively coupled plasma-mass spectrometry (LA-ICP-MS). This new method is showing great potential for measuring trace elements that are sensitive to the differences between sources. The analytical sample is not destroyed or made radioactive by the analysis. More research on this technique will be necessary before it becomes routine, but preliminary work on sources from Yellowstone National Park and the surrounding region has been very positive. This new technique along with XRF and NAA will increase the options for archeologists interested in studying obsidian exchange.

Project title: A Comparison of Northern Elk to Red Deer

Principal investigator: Ms. Teresa Steele
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Stanford, CA 94305-2117

Objective: This study has three objectives: 1) to compare two methods of assigning age to elk specimens, *cementum annuli* and tooth crown height; 2) to compare three methods of mortality profile construction using a sample of known accumulation (elk killed by wolves in 2000); and 3) to determine if mean tooth breadth for a sample can be used as a proxy for population body size. *Cementum annuli* data were collected by park biologists for their own projects, and I measured tooth crown height and breadth and metapodial breadth and depth on the same individuals. The ultimate objective of this research is to develop tools to help understand fauna found in archeology sites. Archeologists need to know how best to determine age on isolated ungulate teeth found in their assemblages. Mortality profiles found in archeology sites provide information about the mode of accumulation of the assemblage, but they are best understood in light of known comparative samples, such as this sample of elk killed by wolves.

Findings: I have completed data collection and am proceeding with analysis. Age classes based on tooth crown heights are similar to cementum annuli ages in younger animals, but crown height reached zero (and well below) before the animals were killed. This indicates that crown height is not a good measure of age in these older individuals. This work will not be written up until the NPS publishes their *cementum annuli* data. The three methods for constructing mortality profiles have been compared using the wolf kill sample, and all methods reconstruct an attritional profile using tooth crown heights. I presented a preliminary version of this work, "A comparison of different methods for analyzing the age structure of archeological samples of hypsodont ungulates," at the Society for American Archeology meetings in New Orleans, Louisiana on April 20, 2001) Currently, I am preparing this research for publication. I have not begun to analyze the tooth breadth data, but I will this autumn.

BOTANY

Project title: Vascular Flora of the Greater Yellowstone Area

Principal investigator: Mr. Erwin Evert
Phone: 847-823-1501
Address: 1476 Tyrell
Park Ridge, IL 60068

Objective: To collect vascular plant specimens as vouchers for distribution maps to be included in the investigator's "Flora of the Greater Yellowstone Area."

Findings: Nine species previously unreported for YNP: *Carex scirpiformis, Antennaria lanata, Eriogonum umbellantun var dicrocephalum, Carex rupestris, Antennaria aromatica, Carex submigricans, C. spectabilis, Dryopteris filix-mas* and *Valeriama acutiloba.*

Project title: Above- and Below Ground Carbon Allocation in Developing and Mature Lodgepole Pine Forests in Yellowstone National Park

Principal investigator: Dr. Dennis Knight
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Department of Botany
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Laramie, WY 82071-3165

Additional investigator(s): Creighton M. Litton, Michael G. Ryan

Objective: Fire and landscape variables interact to produce a mosaic of different vegetation types. The resulting spatial heterogeneity in tree density, herbaceous cover, and species composition that has been observed in Yellowstone National Park will influence primary production and carbon storage for many years. Therefore, to determine the long-term effects of fire on carbon release and storage during succession, we must understand how processes differ among sites as a function of community structure. Our research focuses on the effects of differences in growth-form composition (i.e., relative abundance of trees and herbaceous plants) on above- and below ground carbon dynamics following the 1988 fires in Yellowstone National Park. The general objectives are to answer two questions: 1) How do above- and below ground carbon storage and flux values differ in 12-year-old post-fire stands with different proportions of trees and herbaceous plants? 2) How do above- and below ground carbon storage and flux values in stands burned 12 years ago differ from comparable values in nearby mature forests with similar soils? Answering these questions will enable us to look more holistically at the effects of differences in growth-form composition on carbon allocation across the Yellowstone landscape where lodgepole pine *(Pinus contorta var. latifolia)* is

the dominant species.

Findings: A four month field season during summer 2000 allowed significant progress on our study, including: 1) the location and establishment of all plots; 2) measurement of soil CO2 efflux at four different times; 3) estimation of herbaceous aboveground net primary productivity; 4) estimation of litterfall rates for summer 2000; 5) collection of soil samples for estimating soil organic matter, root carbon pools, and physical properties (i.e., bulk density and texture, plus microbial biomass); 6) collection of stand basal area data for estimation of dead coarse woody root carbon pools; 7) placement of ion exchange resin bags for estimation of N availability; and 8) collection of samples for estimating litter carbon pools. Most of the samples will be analyzed this winter.

Additional field work during 2001 is required. Specifically, we must: 1) sample soil CO2 efflux rates early in the summer when soils are saturated from melting snowpack and soil temperatures are still relatively low; 2) complete data collection for aboveground net primary productivity estimation; 3) collect samples for estimating winter litterfall rates; 4) collect ion-exchange resin bags that were placed in the field during September 2000; and 6) resample soil, litter and root carbon pools to provide estimates of change in these compartments.

Project title: Physiology of Thermotolerant Plants in Yellowstone Park

Principal investigator: Dr. Richard Stout
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Address: Department of Plant Sciences and Plant Pathology
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Montana State University
Bozeman MT 59717

Objective: Our chief objectives for 2000 were to continue to monitor the rhizosphere temperatures of plants growing in several geothermal areas in Yellowstone and to continue a systematic vegetative pattern survey in these areas.

Findings: This year we collected further long-term rhizosphere temperature data regarding the grass species *D. lanuginosum.* These results were in general agreement with results from previous years, that is, the roots of this plant are exposed to temperatures exceeding 40 C for weeks. In addition, we collected vegetative pattern data in selected areas at Amphitheater Springs, 100 Spring Plain and Rabbit Creek. At this time the data is inconclusive regarding correlations between physical factors and species distribution in geothermal areas. We plan to continue this research next year.

Project title: Yellowstone Flora

Principal investigator: Ms. Jennifer Whipple
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Address: National Park Service
P.O. Box 168
Yellowstone NP, WY 82190

Objective: The vascular plant flora of Yellowstone, even though investigated for approximately 120 years, is not completely known. The primary focus of this project is to improve the current knowledge of the flora of the park through in-depth collecting, especially of areas in the park that have not been previously studied. This includes inventory of the occurrence and range of native taxa and also involves the documentation of the arrival and spread of exotic species. In addition, collection of specimens for the Yellowstone herbarium will improve the value of the facility for both NPS personnel and outside researchers.

Findings: Ongoing inventory of vascular plants and collection for the Yellowstone National Park Herbarium. Eleven species of vascular plants previously not reported or not confirmed as occurring within the park were discovered. *Astragalus inflexus Dougl ex. Hook.* [bent milk-vetch], *Orogenia linearifolia Wats.* [linear-leaved orogenia], *Polygonum minimum Wats.* [broadleaf knotweed], *Antennaria lanata (Hook.) Greene* [woolly pussy-toes], and *Townsendia condensata Parry ex Gray var. condensata* were all located or confirmed to occur in the park. These native species are presumed to have been a long-term component of Yellowstone's flora that had been previously overlooked or reported based on material either not collected in Yellowstone or on misidentified material.

Six new exotics to the Yellowstone flora were located within the park. *Ambrosia artemisiifolia L.* [annual ragweed], *Juncus compressus Jacq.* [compressed rush], and *Tamarix chinensis Loureiro [*tamarisk] were located by resource management personnel. In addition, *Arctium lappa L.* [great burdock], *Asparagus officinalis L.* [asparagus], and *Astragalus cicer L.* [chick-pea milk-vetch] were also located within the park. All of these species were either immediately eradicated if possible, or are being actively controlled.

CLIMATOLOGY

Project title: Snow Pack on the Northern Range

Principal investigator: Mr. Phillip Farnes
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Bozeman, MT 59771-0691

Objective: Evaluate variability of snow pack across YNP with emphasis on the northern range and determine sinking depths of different bearing pressures on different snow conditions.

Findings: Data collection is continuing, but data on snow water equivalent will be used in snow modeling efforts across YNP. Data on sinking depths will be analyzed at completion of project and reported in a technical paper. Preliminary results indicate snow accumulation across the northern range follows a predictable pattern but varies by season and year. On sinking depths, more dense snow will support greater bearing pressure.

Project title: Climatic Variation in the Greater Yellowstone Ecosystem: Evaluating the Evidence for Decade to Centennial Variability in Climate

Principal investigator: Dr. Lisa J. Graumlich
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P.O. Box 173490
Bozeman, MT 59717-3490

Additional investigator(s): Mike Pisaric, Lindsey Waggoner, Jeremy Littell, Andy Bunn

Objective: An extended record of climatic variability in the GYE will enhance our understanding of regional patterns and processes. For example, studies of the interactions between climatic variability, fire, and grazing in regulating forest stand structure and composition will be enhanced by longer and more detailed climatic histories of the region. Similarly, research on interactions of fire, climate and geomorphic processes will benefit from better information on climatic trends and variability. Finally, long-term histories of climate can inform the monitoring strategies for assessing the impact of global environmental change on mountain regions

Findings: We continue to develop chronologies for multiple species and monitor real time temperature data at several sites in the park.

ECOLOGY

Project title: Cross-Boundary Plant Invasions in Protected Areas: The West Yellowstone Area

Principal investigator: Dr. Paul Alaback
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Additional investigator(s): Anibal Pauchard

Objective: To 1) determine susceptibility of plant communities to invasion at the landscape scale in the interface between Gallatin NF and Yellowstone NP; 2) determine both forest edge effects on alien plant invasions and biodiversity patterns, contrasting natural and human disturbances; and 3) determine the invasive strategy of *Linaria vulgaris Mill.* at multiple spatial scales in the study area and characterize its potential to invade areas in relation with disturbance types and regimes.

Findings: Our results show that invasions are widely spread across the study area, but are much more prominent in the Gallatin NF. In Yellowstone NP, invasive species occupy disturbed areas along roads and seem to be expanding to more pristine areas such as riparian corridors and naturally disturbed areas. We found significant differences in community patterns in relation to disturbance types. Road edges have the higher numbers of exotic species, followed by clearcut edges. Fire edges have a low number of exotic species. In general, invasive species do not penetrate forest edges. Based on our preliminary results, we conclude that *L. vulgaris* is primarily colonizing old clearcuts and other areas with a disturbed soil layer. *L. vulgaris* patches do not seem to be dying back and their overall population is increasing. We conclude from this preliminary analysis that invasive species in the study area respond both to landscape mechanisms of dispersal along corridors across patch mosaics and site level disturbance factors. While *L. vulgaris* is generally not considered a major species of concern in the northern Rockies regionally, in the unique climatic and geological setting of GYA our data suggests common toadflax has the potential to become a significant problem both inside and outside the park. We will collect more data in the field during the summer 2001 and will conduct other statistical analysis to complete our research project.

Project title: Relating Avian Abundance and Diversity to Human Disturbance Regimes via Hyperspectral Imaging

Principal investigator: Mr. Eric Atkinson
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Address: Marmot's Edge Conservation

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Belgrade, MT 59714

Objective: To 1) identify areas of significant avian abundance diversity within riparian zones on the northern range; 2) identify areas supporting amphibian breeding, foraging, and dispersal activities within riparian zones on the northern range; 3) relate hyperspectral imaging data to patterns of habitat, avian, and herptile abundances and distribution; and 4) relate diversity and abundance patterns to anthropogenic disturbance regimes within and between drainages.

Findings: During our first year of field investigations we sampled 228 point count plots for birds, yielding 3,134 individuals comprising 95 species. The most numerous passerines detected within 100m on point counts were Cliff Swallows *(Petrochelidonpyrrhonata)* owing to their gregarious nature, American Robins *(Turdus migratorius)*, and Savannah Sparrows *(Passerculus sandwichensis)*. Savannah Sparrows were found in sedge- *(Carex sp.)* dominated areas, whereas at the other end of the vegetation gradient, Lincoln's Sparrows *(Melospiza lincolnii)* were consistently found in willow *(Salix spp.)* stands. Thus far, we have identified significant drainage effects upon levels of riparian bird community diversity and abundance. Bird abundance, species richness, and diversity were positively related to willow abundance but showed varying patterns between drainages on the northern range. We are refining measures to more accurately describe species replacements associated with habitat features such as basin characteristics, floodplain size, and riparian shrub height and density. Development of an Index of Biotic Integrity relating specifically to shrub-dependent passerines is ongoing, and assessment of these measures will be done in cooperation with other Yellowstone Ecosystem Studies projects, especially the Hyperspectral Imaging Research. Moreover, these metrics will become environmental parameters for herpetelogical work. These same drainages have been surveyed for amphibians and reptiles identifying one previously unknown boreal toad *(Bufo boreas)* breeding site as well as breeding sites of Columbia spotted frogs *(Rana lutieventris)* and boreal chorus frogs *(Pseudacris maculata)*.

Project title: The Ecological Relationship Between a Rocky Mountain Threatened Species and a Great Plains Agricultural Pest

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Reno, NV 89557

Additional investigator(s): Hillary Robison

Objective: 1) To determine where army cutworm moths *(Euxoa auxiliaris)* (ACMs) originate using microsatellite and mtDNA markers. Pressures on ACM subpopulations, either natural (e.g., weather patterns) or human-caused (e.g., pesticides or habitat loss), may affect moth recruitment and the numbers of adults reaching high elevation sites where they are a critical food source for the threatened grizzly bear

(Ursus arctos horribilis). 2) To determine if ACMs harbor agricultural pesticides in their tissues. Resulting magnification in grizzly bears that forage heavily on moths may have detrimental physiological or developmental side effects. 3) To determine whether ACMs from different Great Plains origins are interbreeding in high elevation sites prior to their return to agricultural areas. If ACM subpopulations do not interbreed, unfavorable conditions in specific Great Plains areas may impact moth numbers in high elevation.

Findings: This project is ongoing.

Project title: Effects of Fires on Ecology of Coyotes in Yellowstone National Park: Baseline Succeeding Wolf Recovery

Principal investigator: Dr. Robert Crabtree
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Email: crabtree@yellowstone.org
Address: P.O. Box 6640
Bozeman, MT 59771

Additional investigator(s): Jennifer Sheldon, David Bopp, Paul Moorcroft, Chris Wilmers

Objective: Document long-term effects of the 1988 fires on the population dynamics and behavioral ecology of coyotes. Document the impacts of wolf restoration on coyote population and behavioral ecology including effects of coyote prey and competitor species. Continue long-term monitoring of coyote populations by adhering to those objectives listed in previous reports and peer-reviewed publications.

Findings: This project is beginning year twelve and is in Phase II: Wolf colonization period. A variety of significant behavioral and demographic effects of wolves on coyotes continue to occur since the release of wolves in 1995. The direct effects of fires on coyotes continue to be insignificant, but indirect effects on the small mammal prey base continue. Currently (2001), thirty-five resident adult coyotes occupy the Lamar Valley study area, and 20 occupy the Blacktail Plateau study area. In the year 2000, the Druid wolf pack expanded its range to include what was formerly a buffer zone between two neighboring wolf packs, and the coyote population in this zone, formerly stable, was reduced by 50 percent, with a concomitant acceleration of alpha pair turnover. Due to the lack of consumable biomass after the larger wolf packs finish their first feeding, coyote use of wolf-killed carcasses has decreased markedly in 2000.

Project title: Landscape Use by Elk During Winter on Yellowstone's Northern Range

Principal investigator: Dr. Robert Crabtree
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Additional investigator(s): David Bopp, Paul Moorcroft, Phil Farnes

Objective: The objectives of this study were to document winter patterns of landscape use by Yellowstone northern range elk; measure elk feeding activity (as indexed by number of feeding craters); quantify snowpack characteristics; and examine how these and other landscape and habitat features influence elk foraging locations. How does snow affect the distribution of elk during winter on Yellowstone's northern range? What other factors, such as winter temperature, forage, and predator/prey density, are affecting their distribution?

Findings: We measured site and snowpack characteristics, elk *(Cervus elaphus)* feeding crater densities and morphometry, and elk numbers in the Lamar River Valley and the Blacktail Plateau on the northern range of Yellowstone National Park, Wyoming. We conducted the study over three winters, 1992-93 to 1994-95, but the main sampling effort occurred over four monthly sample periods in year one. Snow depth (SD), snow water equivalent (SWE) and snow resistance to horizontal movement and vertical penetration all increased steadily over the winter. The mean (SD) feeding crater diameter and depth was 118 (37) cm and 34 (11) cm, respectively, and both were positively correlated with snow depth. The mean (SD) crater volume was 385 (321) l, and the mean (SD) mass of snow excavated from a crater was 82 (72) kg. Nonwoody plants (grasses, sedges and forbs) were the primary browse item in 90 percent of the craters. The highest aerial elk counts were observed in early- to mid-January, and counts declined substantially and steadily after January 29. At this time, mean snow depth was about 50 cm and mean SWE was about 12 cm. The mean number of new craters on a plot showed a significant, negative association with snow depth, SWE and booted-foot sinking depth. We used the sum of craters on a plot across all four sample periods as an index of winter-long feeding activity. Elevation and habitat type were the best site characteristics for differentiating plots in regard to winter-long use. Summed craters were negatively associated with elevation, and the habitat type with the highest summed craters was tufted hairgrass/sedge. Only about 5 percent of plots that had craters had areal crater coverage in excess of 14 percent, with a maximum of 23 percent coverage, suggesting that snow disturbance associated with cratering activity may inhibit elk foraging. We are preparing manuscripts for publication and are also preparing for field work starting November 2001.

Project title: Specificity in Ectomycorrhizal Symbioses

Principal investigator: Dr. Ken Cullings
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Address: NASA Ames Research Center
MS239-4
Moffett Field, CA 94035

Objective: To determine patterns of plant/ectomycorrhizal fungal interaction in multiple forest types, and determine effects of disturbance on these systems.

Findings: 1) no support for specificity; 2) clear patterns of seasonal variation; 3) significant effects of defoliation of pine on mycorrhizae of both pine and spruce; 4) significant effects of litter addition on mycorrhizal community structure in pure pine; 5) significant effects of litter removal on mycorrhizal community structure in mixed pine/spruce; and 6) significant differences between effects of clear-cutting and natural

burn in pure lodgepole pine.

Project title: Molecular Assessment of Microbial Communities in Hot Spring Structures and Their Responses to Light Manipulation

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MS239-4
Moffett Field, CA 94035

Additional investigator(s): Dr. Evan Lau

Objective: To 1) determine patterns of evolution of cyanobacteria forming stromatolites in lithifying mats in hot springs; 2) determine effects of light and temperature gradients on these communities; and 3) Determine if stromatolite morphology is related to cyanobacterial community structure in mats.

Findings: Sampling along thermal gradients has been completed, as has sampling of mat vs. stromatolite and of different stromatolite morphs. DNA analysis is underway. Initial results indicate that cyanobacteria forming these modern stromatolites, that are analogs to 3.5 billion year old fossils, have a single phylogenetic origin.

Project title: Habitat Requirements and Evolution of *Agrostis rossiae Vasey,* a Grass Endemic to Thermal Soils in Yellowstone National Park

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Web site: http://www.agrostis.topcities.com/
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Additional investigator(s): Michael Tercek

Objective: To 1) use genetic markers to determine the phylogenetic relationship between *Agrostis rossiae* and closely related congeners, determine whether *A. rossiae* is a valid, monophyletic taxon or an ecotype of other co-occuring *Agrostis* species; 2) explain the geographic distribution of *A. rossiae* in terms of ecological variables; determine the reasons for its endemism; and 3) Calculate levels of gene flow between *A. rossiae* populations and between *A. rossiae* and co-occurring congeners.

Findings: 1) Genetics samples of 10 *Agrostis* species and over 20 *A. rossiae* populations were collected dur-

ing Spring/Summer 2000. RAPDS primer screening has been completed and data collection is underway. 2) Field measurements of soil temperature, moisture, and chemical composition have prompted greenhouse experiments, which are currently underway. Cross-pollination and self-pollination experiments are also being conducted. 3) Common garden experiments show that *A. rossiae* retains its distinct morphology under a wide variety of growing conditions.

Project title: Browsing Phenolgy of Willows, Cottonwood and Aspen on the Northern Range, Yellowstone National Park

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Additional investigator(s): YES personnel as assigned

Objective: The purpose of this project is to document variation in the timing of winter browsing of woody riparian species and relate it to climatic conditions.

Findings: During the first winter it appears that little browsing occurred until late January/early February, then most of the stems were eaten. One stand was not touched and several inches of last summer's growth were left on most stands. At one site it was noted that many branches were bitten off and left on the ground. It was also decided that a slight change in the observation protocol would provide more information. An estimate of percentage of twigs browsed will be recorded for each of 5 to 10 subplots at each site using the common 6 class system of less than 5 percent, 5-25 percent , 25-50 percent, 50-75 percent, 75-95 percent, greater than 95 percent. This method will be evaluated this winter.

Project title: Assessing Ecosystem Integrity: An Approach to Modeling Energy Flow

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Additional investigator(s): Diane Ashton, Ken Cummins, Peggy Wilzbach

Objective: To determine secondary invertebrate production in wetlands of the Lamar Valley and develop methods for modeling production.

Findings: In 2000, field studies were limited to one visit. During this visit data were collected on respira-

tion rates of six species of aquatic invertebrates inhabiting wetlands. These data will be used in modeling the energetics of these species.

Project title: The Sustainability of Grazing Ecosystems

Principal investigator: Dr. Douglas Frank
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Additional investigator(s): Peter Groffman

Objective: To measure the effects of grazing ungulates on aboveground and belowground production at diverse grassland sites on winter, transitional, and summer ranges.

Findings: 1) In 1999, grazers increased both shoot and root production. 2) Ungulates increased the fine grain spatial heterogeneity of soil N content and N mineralization. c In a growth chamber study, defoliating a common Yellowstone grass, Poa pratensis, increased root carbon exudation, rhizospheric microbial biomass, microbial activity, and N mineralization. These effects in the rhizosphere were associated with increased N uptake, plant N content, and photosynthetic rates in clipped plants.

Project title: Yellowstone Science and Technology Trip

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Additional investigator(s): Travis Muirbrook, Kerry Lambert

Objective: The goal of this experience is to put students into a natural setting that will stimulate their curiosity and excitement. Yellowstone National Park provides the ideal setting to provide students with abundant opportunities to observe and study numerous biological and geological processes. In addition, the students will be provided with cutting-edge portable technology to gather, measure and record scientific observations. The students' E-mate computers provide rd processing, spreadsheet, and drawing capabilities, which are all ideal for recording field data. Yellowstone provides incredible opportunities for observation of various large mammals in their natural environment, and is one of the few natural laboratories in North America where interactions between predators and prey can still be observed, with little or no impact on their natural behavior. Students will use the portable computers to record behavioral observations in the field as they occur. In addition, the E-Mates are equipped with number of probes that can be

used to measure temperature, pH, and dissolved oxygen. These provide data from the numerous thermal features and water bodies in the park. Video and Quick take clips will also be used on the trip to record students' experiences. Upon their return, the students will take their recorded data and complete a scientific summary paper. Students will use video and still shots in addition to their experiences to construct a web page of their trip to be posted on the Internet.

Findings: The only thing collected from the park will be the water data and wildlife observation data that the students will use to prepare a report of their trip. In addition, photography and video will be used to document the student experiences.

Project title: Habitat Use, Diet Quality and Composition of a Pronghorn Population in Yellowstone

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Additional investigator(s): Vanna Boccadori

Objective: To develop robust predictive models that describe how pronghorn use winter and spring habitats and predict pronghorn habitat use under varying environmental conditions. A second objective is to evaluate the nutritional content of pronghorn diets from known botanical diet compositions using fecal analyses, and nutrient consumption of herbages determined from published values. A third objective is to evaluate the parameters of habitat use and diet composition and quality of YNP's declining pronghorn population within two frames of reference: in the context of other pronghorn populations, both healthy and of low productivity, in comparable studies; and in the context of findings from studies done on this population of pronghorn between 1967-1970 and 1986-1988. An assessment of the current pronghorn winter range will be investigated regarding changes in management, land use, ownership, and quality and quantity of vegetation communities to provide context for the latter comparison. A fourth objective is to describe seasonal home ranges and migration routes, and to provide basic demographic information.

Findings: 1504 observations were made on collared pronghorn during the period of June 1, 1999 and August 31, 2000. The majority of observations occurred during January to March 2000 when the pronghorn were concentrated on the winter range. Prior to and following these months the pronghorn displayed greater mobility over a larger extent of landscape. More time was required to locate pronghorn during these periods. Five pronghorn were collared in March 2000. Six collared does died during the year 2000. The majority of observations made during the period of November 1999 to March 2000 occurred within YNP, and extended from Devil's Slide to Slide Lake on the Old Gardiner Road. Spring migration occurred during April and May 2000. One collared doe migrated out of the park during the spring to the area around Rock Creek in Tom Miner Basin, and currently is still there. Nineteen collared does migrated from the winter range to points from McMinn Bench on the west to Lamar Valley to the east, Specimen Ridge

on the south to the Yellowstone and Lamar Rivers to the north. Seven collared does remained on the winter range throughout the summer. All collared does except the doe at Rock Creek returned to the winter range by November 2000.

The use of vegetation community-type by collared pronghorn from November, 1999 through July 2000 were quantified. The data were obtained from visual observations of the community-type the collared doe was in at the time of observation. Observations occurred more frequently in grass-dominated communities throughout all months except May 2000, when the majority of relocations occurred in sagebrush communities.

The second field season resumed in November 2000. Data-collection methods employed last season have proven effective and will be used again during the current field season. The research is projected to be completed by December 31, 2001.

Project title: Ungulate Carrion Impact on Plant Community Composition and Nutrient Cycling

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Additional investigator(s): Christopher Wilmers

Objective: To determine whether wolf-killed ungulate carcasses are having an impact on plant community composition and nutrient cycling.

Findings: We determined that the sample size required to get a significant result was unattainable. We initially marked 5 rumen piles which all turned out to be in different plant cover types. Wolf kills occur in approximately 10 different plant cover types, so in order to do this study effectively we estimated that we would need at least 10 samples in each of the 10 cover types, which we did not feel we could achieve.

Project title: Development of an Empirical Model for Predicting the Stream Invertebrate Fauna of the Greater Yellowstone Ecosystem: a pilot study

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Additional investigator(s): Trey Simmons

Objective: The primary objective of this study is to develop an empirical model that accurately predicts the

expected invertebrate fauna in streams of the Greater Yellowstone Ecosystem. Models of this type can be used to assess the biological integrity of streams that may be impacted by anthropogenic activity. As predictive models have generally been applied across relatively large spatial scales, we were interested in assessing how they might perform at a relatively small scale, where latitudinal and elevational differences are minimal. An additional objective was to assess whether detailed temperature data improved the performance of these models.

Findings: We collected benthic macroinvertebrates (both quantitative fixed area and qualitative timed samples), periphyton, physical data, and three months of continuous temperature data from 31 streams and rivers in Yellowstone National Park. Temperature data were collected from late June to late September. Sampling and physical data collection were conducted in August and early September. Invertebrates and periphyton are currently in the process of taxonomic identification; however, preliminary results indicate that the small scale model performs reasonably well, and that performance is improved by the inclusion of detailed temperature data.

Project title: Cougar-Wolf Interactions In Yellowstone National Park: Competition, Demographics, And Spatial Relationships

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Additional investigator(s): Toni K. Ruth, Howard B. Quigley

Objective: To 1) document the characteristics of the cougar population, including population size, survival, cause-specific mortality, natality, and make comparisons with analogous parameters made prior to wolf restoration (Phase I data); 2) assess the effects of cougars on elk and mule deer populations as influenced by the presence of wolves; 3) assess competition and resource partitioning between cougars and wolves by comparing the species' spatial and temporal habitat use patterns and prey utilization characteristics; 4) quantify spatial and temporal interactions between cougars and wolves; and 5) communicate research findings to state and federal agencies and the general public through annual technical reports, research updates, and presentations.

Findings: Hornocker Wildlife Institute (HWI) personnel captured and radio-collared a total of 36 cougars in and adjacent to areas used by three wolf packs on the Northern Yellowstone Study Area (NYSA), Montana, and Wyoming. A sample of 3 to 10 radio-collared wolves was maintained within each wolf pack by the Yellowstone Wolf Restoration Program. Researchers associated with both HWI and Yellowstone National Park (YNP) conducted aerial and ground monitoring of radio-instrumented animals. To conduct winter cougar sign surveys and provide and estimate of cougar populations size, field crews searched 1589 km of track transect during winter 1998-1999 and 1208 km during winter 1999-2000. A minimum of

21-22 adult and subadult cougars were present on the NYSA during the 1998-2000 winter seasons. Ten resident adults (3 male; 7 female) and 5 kittens in 3 family groups are currently being monitored on the NYSA. Six adult females produced 8 litters of 2-4 kittens, resulting in 21 offspring documented during March 1998-August 2000. Eight cougar deaths were documented. All four of female F107's kittens were killed by the Druid wolf pack in two separate events occurring near an elk killed by F107. Three cougars were killed by other cougars and one cougar kitten was killed by a black bear. Eleven cougars have dispersed to areas adjacent to and beyond the NYSA. One-hundred thirty-six positive and probable cougar kills were documented. Prey included 98 elk, 22 mule deer, one bighorn sheep, one antelope, five coyotes, four porcupines, one red squirrel, and one golden eagle. Seven predation sequences of 26 to 35 consecutive days resulted in a mean predation rate of 7.4 days per ungulate kill for the seven cougars sampled. Twenty-seven percent of cougar-killed ungulate prey were scavenged or usurped by other carnivores. We documented visits to 122 cougar kills by wolves (4.9 percent), grizzly bears (4.9 percent), black bears (9.0 percent), coyotes (14.8 percent), foxes (1.6 percent), and other cougars (0.8 percent). Cougars were displaced from kills by wolves in 5 of 6 documented visits.

Project title: Determining Forage Availability and Habitat Use Patterns for Bison in Hayden Valley of Yellowstone National Park

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Additional investigator(s): Tom Olenicki, Peter Gogan, Robert Garrott

Objective: 1) Describe seasonal bison habitat use patterns and factors that influence these patterns in the Hayden Valley of Yellowstone National Park. 2) Identify interactions between bison and vegetation. 3) Develop a monitoring strategy to track changes in vegetation due to ungulate herbivory. 4) Provide baseline data for models of ungulate-vegetation relationships in the Hayden Valley.

Findings: During the 2000 field season we continued monitoring bison distribution and forage utilization. We obtained ground calibrations to develop forage biomass estimates from multispectral imagery for the third year. We measured utilization via exclosure cages for the last of three years and removed all exclosure material from Yellowstone. GPS technology was used to delineate fine scale foraging patterns of bison. Data collected in this and the previous two field seasons will allow us to make spatially explicit estimates of biomass in individual plant communities, produce spatially explicit temporal estimates of plant phenology, and determine how biomass, phenology, or community species composition interact to drive bison foraging patterns. Brief field visits will be necessary in summer 2001 to refine the vegetation map and complete species composition information on vegetation in the Hayden Valley.

Project title: The Dietary and Foraging Ecology of the Yellowstone River Otter: An Umbrella Species for Aquatic Systems

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Additional investigator(s): Bill Wengeler, Nathan Varley

Objective: We propose to clarify the ecological role of native vs. non-native fish species on Yellowstone river otters by comparing otter populations on two lake systems in YNP: Yellowstone Lake, where the native cutthroat population is still healthy, and the Lewis-Shoshone Lake complex in which non-native fish, primarily lake trout and brown trout, predominate. Specifically, we are characterizing river otter diets through fecal analysis. Furthermore, we are employing stable isotope analysis to determine the trophic interactions among otters and their prey. The data generated by this research will help assess the consequences of the lake trout invasion on a predator whose existence is firmly linked to aquatic systems and may prove critical to the future management of Yellowstone waters.

Findings: Collection of river otter fecal specimens continued this year. In addition, representative fish and invertebrates were collected from Yellowstone and Lewis Lakes for stable isotope analysis. Hair clipped from mounted river otters was collected for the same purpose. Initial stable isotope analysis has been completed. We are currently analyzing fecal samples in the lab and characterizing otter diet by the frequency of occurrence of various prey items. Preliminary data suggest that there is a significant difference in otters' dietary reliance upon trout between the two lake systems. Initial examination of lake and cutthroat trout otoliths indicates that it is likely possible to distinguish the two species on the basis of otolith morphology.

Project title: '96 Field Course: Forest Ecology and Geology of the Yellowstone Country

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Additional investigator(s): Dr. Martin Knoll

Objective: Primary forest objectives are to 1) identify stands representative of the various forest habitat and cover types described by Despain (1990); 2) establish permanent plots within each stand; 3) develop a database of stand characteristics such as tree species composition, height, age, and understory composition

for each plot; and 4) establish transects to investigate species gradients in relationship to thermal features. The plots will be used to illustrate the concept of habitat typing as applied to Yellowstone forests. Primary geology objectives are to 1) study temperature and pH gradients in Yellowstone Lake and representative hot springs; 2) characterize thermal features representative of different regions of the park; and 3) identify rock types characteristic of the major volcanic episodes in Yellowstone NP. The field course is taught during alternate years.

Findings: No activity was conducted this report year.

Project title: Non-Native Plant Monitoring

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Objective: To complete several non-native monitoring projects initiated in Yellowstone since 1995. That included a re-survey of 60 sites established across the park in 1995 and 1996 that were used to assess native and non-native vegetation. Vegetation analyses were done on quadrants at each of those sites and each of the 60 sites was evaluated twice during the season. Foot surveys to evaluate non-native vegetation were also done along 0.25 mile long strips at each site and drive surveys were done on the intervening 5-mile long road sections between sites too. In addition, U.S. 191 was re-surveyed for non-native plants to compare with the results of a study done several years ago.

Findings: The data set produced during this past season is still undergoing evaluation. A technical report of the work done on U.S. 191 is basically done but requires some verification work/fine tuning before submission. The data for the 0.25 mile long park-wide foot surveys at all 60 sites has been entered into spreadsheets as well as the drive survey information for the 5-mile long study sections. Analysis/writing will continue through the spring/summer until completion with technical report submittal to the appropriate offices. A draft manuscript for the entire project will be prepared in collaboration with Tom Olliff and Craig McClure (NPS).

Project title: Study of the Effects of the 1988 Wildfires on Yellowstone Stream Ecosystems

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Additional investigator(s): Andrew Marcus, Robert Crabtree

Objective: The overall objective of this study is to separate the early from the delayed effects of wildfire on stream ecosystems in Yellowstone National Park. Specific goals include documenting changes in stream habitat and biota each successive year following the 1988 wildfires, thus providing a basis for predicting and evaluating the subsequent long-term changes. Few streams greater than 4th order in size were substantially affected by the fires, and this study focuses on stream of 1st through 4th order. To increase the breadth and precision of the study and to provide more general conclusions, each size class (order) is represented by four or five streams, as well as by at least one reference stream that was not affected by the fires. Since 1993 (except for 1998), the research has been limited to streams in the Cache Creek drainage basin and immediately adjacent reference streams due to financial constraints.

Findings: Although the effects of fire were evident in early years of this study, the streams could be characterized as being largely on a "fast recovery track." However, 1991 and 1994 were marked by runoff events that caused substantial alteration of physical habitat in the streams in burned watersheds, particularly those in moderate to steep gradients. Even greater physical alterations occurred in 1995 (and probably in 1996) and were evident again in 1997. The dramatic changes in 1995-1997 are associated with a general increase in precipitation in those years. Disturbances such as these are reflected in declines in the biotic components of the stream and serve as important "resets" in the recovery process. However, as was evident from examination of one of our reference streams (Amphitheater), some of those differences are more a response to a change in annual weather conditions than to fire per se.

The most dramatic differences in the Yellowstone post-fire streams have been in physical alterations of the stream channel and biological restructuring of communities. Channel morphology is different in burned, large order streams, as bank width is larger than in the large unburned stream. Substrate particle size is different also between comparable-size burned and unburned streams. The smaller Cache sites are receiving more fine sediment from surface erosion than the comparable unburned stream. This pattern is not seen in the larger burned streams perhaps because increased stream discharge flushes the fine sediment downstream. Biological patterns among burned and unburned streams are complex, but differences have emerged in the periphyton communities and in the restructuring and densities of macroinvertebrate communities. Chlorophyll and ash-free dry mass (both measures of primary production) are significantly higher in small burned streams. This increase in primary production is predicted as canopy cover is removed and more available light is converted for use in photosynthesis. Increases in primary production in first and second order burned streams may be responsible for the dramatic shift from mayflies, caddisflies, and stoneflies (EPT taxa) to midges. In the first ten post-fire years increases in Baetis (a mayfly) and midges were seen, as these taxa are more invasive and disturbance-adapted. Macroinvertebrate results from 2000 will aid in determining whether there is a relationship between high chlorophyll a concentrations and midge density at the expense of EPT taxa, as chlorophyll a concentrations were double in 2000 than those in 1999. If this relationship holds we expect to see midge densities the same or higher for the smaller Cache sites than those found in 1999. The percent EPT taxa was consistently 70 to 80 percent of the total taxa in unburned and large burned streams for 1998 and 1999. A large difference was found however, in the percent Baetis which increased nine times that found in 1998 in the large burned streams. Further research will aid in further establishing these mid- to long- term community patterns in burned streams.

Project title: The Effect of Environmental Variability on Grizzly Bear Habitat Use

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Additional investigator(s): Dr. Robert Garrott, Dr. Charles Schwartz

Objective: The overall design of this project is to utilize existing data, expertise and newly collected data from advanced technologies to evaluate the impact of anthropomorphic influences on grizzly bear (Ursus arctos horribilis) habitat selection. Initially, this study will have three areas of emphasis: 1) to evaluate potential relationships between habitat use and road density; 2) to evaluate potential relationships between habitat use intensity and types of human activity; and 3) to evaluate potential relationships between habitat selection and land management status.

Findings: During the 2000 field season the Interagency Grizzly Bear Study Team and Wyoming Game and Fish were able to instrument 15 bears for this project. Of the 15 grizzly bears collared, 6 were adult females, 2 were sub-adult females, 5 were adult males and 2 were sub-adult males. The first collar was deployed on May 3, 2000 and the last collar was deployed on September 22, 2000. These collars have a programmable duty cycle that we set to attempt a location collection every 210 minutes. The collars will power down on November 15, 2000 and come back on April 15, 2001. The collars also have a remote release mechanism which will automatically release the collar in the middle of summer 2001. Collars will be collected and data downloaded by the researchers. Also during the 2000 field season we collected the remainder of down collars for the 1999 field season, updated ancillary biophysical data sets and began preliminary data analysis. Next year the researchers plan to use this same collar technology and attempt to deploy the 14 more collars. In addition to the collection information on grizzly bears the researchers will continue preliminary data analysis, test the effect of canopy on location acquisition and update the current grizzly bear habitat coverages and other biophysical data layers with available data.

Project title: Developing Effective Ecological Indicators for Watershed Analysis

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Additional investigator(s): Andrew Marcus, Rick Lawrence, Wayne Minshall

Objective: This study is designed to develop improved indicators and innovative techniques for assisting and monitoring ecological integrity at the watershed level in the western United States. Its specific objectives are to develop practical, scientifically valid indicators that 1) span multiple resource categories; 2) are relatively scale independent; 3) address different levels of biological organization; 4) can be rapidly and cost-effectively monitored by remote sensing; and 5) are sensitive to a broad range of anthropogenic and natural environmental stressors.

Findings: None to report.

Project title: Ecology of Selected Habitats in Yellowstone National Park

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Additional investigator(s): Jeff Schloss

Objective: To 1) quantify the age distribution and density of lodgepole pine in unburned areas and pine recovery in 1988 burned and 1984 blowdown areas; 2) evaluate the influence of thermal runoff on aquatic macroinvertebrates in streams draining geyser basins and compare them to streams lacking thermal runoff that were sampled outside of the park; and 3) observe the adaptations of bacteria, algae, and other life to the diverse hydrothermal features unique to YNP.

Findings: We found that lodgepole pine occurs in burned areas at densities nearly 10 times those in unburned and blowdown areas. Most trees in the burned areas were approximately 11 years old with 3-9 cm main stem diameters. The Firehole River was significantly warmer (26C) than streams at comparable elevations outside the park (e.g., the Tongue River in the Bighorn Mountains [18C]) and harbored some invertebrates (e.g., amphipod crustaceans and gastropod mollusks) that streams without thermal runoff did not. At West Thumb Geyser Basin, as has been previously published, distributions of bacteria, algae, and cyanobacteria were related to water temperature, as determined by visible color of the microbial mats. These results are intended for educational use only and not for scientific discovery; nor are they intended for publication.

Project title: Multi-Trophic Level Responses to the Addition of a Top Carnivore

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Additional investigator(s): Dr. L. David Mech, Dr. Mark Boyce, Dr. Evelyn Merrill, Dr. Doug Smith

Objective: This study examines the ecological changes associated with re-establishment of wolves in Yellowstone National Park in 1995 and 1996. Species representing three important trophic levels—wolves, elk, and woody vegetation—are the focus of the research. The specific areas of interest are: 1) spatial and temporal patterns of abundance of the newly introduced top carnivore (gray wolf), the dominant herbivore (elk), and woody vegetation on YNP's northern range and 2) mechanisms underlying trophic dynamics, especially predation rate of wolves and herbivory use by elk on woody vegetation.

Findings: On March 15 and 16, 2000, 45 cow elk were captured via helicopter netgunning on YNP's northern range. The elk were fitted with mortality-sensing radio collars, and a full range of physiological samples was obtained from each elk, including an incisor for aging by *cementum annuli*. Since capture the elk have been monitored several times per week by ground and/or air to examine habitat associations and temporal/spatial patterns. Beginning in late June, the radiocollared elk were tracked every 7-14 days using aerial telemetry. During the summer, these animals ranged throughout the Quadrant, Buffalo Plateau, Mirror Plateau, Cache Creek, Cold/Mist Creeks, and Heart Lake regions. Fall migration occurred between mid-October and early November, during which time all of the tagged elk returned to the northern range. Monitoring of the study animals will continue through 2001 and preliminary analysis of habitat selection has begun. Final habitat selection models are expected to be available in April 2002.

Four of the 45 elk died in 2000 of the following causes: cougar-kill, wolf-kill, unknown predation, and uncertain cause of death (possible fall). Two of the four elk that died may have been hampered by capture-related injuries. Wolf monitoring occurred on a continual basis with intensified efforts during the two winter study periods of Nov/Dec and March. As of year-end 2000, an estimated 168 wolves in 16 packs inhabited the Yellowstone ecosystem. Of this total, 5 packs, or 72 wolves occupied the northern range. As expected, the northern Yellowstone elk herd is providing most of the prey consumed by Yellowstone wolves. The northern herd population reconstruction effort is underway. Elk mortality data has been obtained from the wolf project database (Doug Smith), the Gardiner late hunts (Tom Lemke), and from winterkill transect surveys carried out by a cooperative team (Peter Gogan, BRD-USGS). A methodology for combining these sources of data to obtain a final minimum number alive (MNA) estimate of the herd is currently being developed.

Project title: Aspen Regeneration in Northern Yellowstone National Park

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Additional investigator(s): Eric J. Larsen, Roy A. Renkin, Douglas W. Smith

Objective: Our objective was to measure aspen regenerative success inside and outside of established wolf pack territories on YNP's northern range. Based on trophic cascades theory, we hypothesized that wolves may displace elk from some areas of the northern range, thus allowing more robust aspen regeneration in areas of higher wolf presence. Using radio telemetry data on wolves and a fixed kernel estimate, we delineated polygons representing high-use winter activity areas of the Leopold, Rose Creek, and Druid wolf packs. These polygons were then overlaid onto a map of northern range aspen stands. Aspen stands were divided into two groups, those within polygons representing a high density of wolf telemetry locations (less than 50 percent fixed kernel estimate) and aspen stands in areas of lower densities of wolf telemetry locations (greater than 75 percent fixed kernel estimate). Permanent 1x20 m belt transects (plots) were established in the aspen stands, marked with both a metal identification tag on a large-stemmed aspen tree and nails in the ground at 3,5,10,and 20 m from the starting point.

Findings: Field data were collected from 112 plots in northern range aspen stands. Differentially corrected GPS readings were obtained from each stand. Aspen overstory density and diameter at breast height (DBH) were recorded. Sucker density, heights, and whether the suckers had been browsed the previous winter were recorded. The number of elk pellet groups was recorded for each plot. The generalized habitat type of each aspen stand was recorded using the categories of mesic upland steppe, xeric upland steppe, and wet meadow/riparian. The aspect, slope, elevation, and recent fire history of each stand were recorded. We have summarized our findings in a manuscript that is currently in review for publication and we plan on collecting new data from our permanent plots during the summer of 2001.

Project title: Causes and Consequences of Alternative Successional Trajectories Following the 1988 Yellowstone Fires

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Additional investigator(s): Monica G. Turner, Gerald A. Tuskan, Dennis H. Knight

Objective: To 1) predict and map the early successional pathways of areas burned in 1988 on the basis of percent serotinous lodgepole pine trees, size of burned patch, and local severity of fire; 2) map percentage of serotinous trees across the landscape; 3) measure aboveground net primary productivity and leaf area index in stands representing different initial pathways of plant succession following the Yellowstone fires of 1988; and 4) re-sample plant cover and density in the permanent plots established in 1990 within nine different patches of 1988 crown fire.

Findings: 1) We obtained 1:30,000 color aerial photos of the entire Park in August 1998. Approximately

10 percent of the area that burned in 1988 now supports very high-density stands of 10-year old lodgepole pine trees (greater than 50,000 stems/ha); 10 percent supports very low-density lodgepole pine (less than 100 stems/ha); and the remaining burned area has stands of intermediate tree density. 2) Initial sampling of percent serenity indicated highest percentages at lower elevations in the west-central portion of Yellowstone NP, and lowest percentages in high-elevation forests in the central and eastern portions of the park. 3) Aboveground net primary productivity (ANPP) and leaf area index (LAI) were measured in 1999 in 88 stands that had burned in 1988. These fundamental measures of ecosystem function varied with sapling density, ranging from 0.9 - 12.6 Mg/ha/yr (ANPP) and from 0.03 - 4.6 m2/m2 (LAI).

Project title: Willow Persistence in Yellowstone National Park: Interactive Effects of Climate, Hydrology and Herbivory

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Midcontinent Ecological Science Center
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Additional investigator(s): Tom Hobb, Linda Zeigenfuss

Objective: To provide some baseline data for developing a research proposal on the effects of population limitation of elk by wolves on willow communities on the northern winter range.

Findings: Study sites that were part of studies conducted by F. J. Singer from 1988-1992 were revisited and we collected data on willow consumption by ungulates and previous year's willow production and growth. This information was incorporated into research proposals recently submitted to the park and National Science Foundation.

Project title: Yellowstone Gray Wolf Restoration Project

Principal investigator: Dr. Douglas Smith
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YCR
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Objective: Restoring wolves *(Canis lupus)* to areas where they were eradicated has been an issue for well over 20 years. Yellowstone National Park, with its plentiful ungulates and large protected ecosystem, has figured prominently in all discussions concerning wolf restoration. For many years it was hoped that

wolves would naturally recolonize the Greater Yellowstone Ecosystem, as they did northwestern Montana around Glacier National Park. However, after in-depth considerations of all aspects related to wolf recovery in the northern Rocky Mountains it was decided to reintroduce wolves to the Yellowstone Ecosystem as part of a program to also return self-sustaining wolf populations to suitable habitat in northwestern Montana and central Idaho. The goal is to have 10 wolf packs (about 100 wolves total) in each of these three areas. When 10 packs have produced wolf pups for three consecutive years in each of the three areas, the gray wolf will be proposed for removal from the list of endangered species and managed as a resident species by the states of Montana, Wyoming, and Idaho within their jurisdiction.

Much work remains after wolves are reintroduced to ensure that the animals form a fully restored and self-sustaining population. The wolves must be closely monitored to respond to any problems that may arise, to determine if and when the goal of a self-sustaining population of wolves has been reached, and to learn about their effects on other ecosystem inhabitants. Accordingly, this plan describes the actions necessary to document establishment of packs, adult wolf survival and mortality, population dispersal and distribution, wolf prey selection and predation rates, and den site location and pup survival.

Findings: At the end of 2000, at least 165 wolves in 16 packs were present in the Greater Yellowstone Area (GYA). Eleven of these packs were considered a breeding pair by the definition established by the U.S. Fish and Wildlife Service (a breeding male and female with two pups that survive to December 31). Eight packs (~126 wolves) reside within YNP, and 8 packs (two in Montana and six in Wyoming) range entirely outside the park (~39-43 wolves). Removal of wolves from the endangered species list requires 30 breeding pairs distributed throughout the three recovery areas (GYA, central Idaho, and northwest Montana) for three successive years. In 2000, there were 25 breeding pairs in the northern Rocky Mountains; therefore 2000 was not a countdown year.

Sixty-nine to 78 pups survived to December 31, 2000. Fifty-five to 60 of these pups were born in YNP. Pup survival was higher in YNP in 2000 (55 of 69, 80 percent) than in 1999 (18 of 40, 45 percent). Parvovirus possibly explained the low survival in 1999. All captured wolves in 1999 and 2000 tested positive for the disease, and mortality occurred at a time (post-weaning) when pups are maximally vulnerable to infection. To positively identify mortality due to parvovirus a dead pup must be collected, and thus far no such carcass has been retrieved.

Fifteen litters were born to eleven breeding pairs. Two packs had more than one litter, the Druid Peak pack had at least three, and the Rose Creek pack had two. Because a breeding pair is only counted if a male and a female successfully raise two pups to December 31, and in both of these packs one male bred more than one female, each pack counts as one breeding pair. Litter size ranged from 4-10 and averaged 6.4 (N = 12).

Project title: Biogeochemical Interactions at Environmental Interfaces (Mercury dynamics in aquatic ecosystems of Yellowstone National Park)

Principal investigator: Dr. Robert Striegl
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National Research Program
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Denver CO 80225-0046

Additional investigator(s): David Krabbenhoft, Kimberly Wickland, Mark Dornblaser

Objective: The objectives of this study are to understand the dynamics of mercury and methyl-mercury in ecosystems of YNP, with particular emphasis on Nymph Lake and areas in the vicinity of Norris Geyser Basin.

Findings: Nymph Lake and nearby ponds, streams, and wetlands of YNP have the highest known natural concentrations of dissolved total-mercury and methyl-mercury measured in any aquatic ecosystem anywhere. Impacts on biota living in the area are not yet known.

Project title: The Behavioral–Ecological Role of Wolf Howling

Principal investigator: Dr. John Theberge
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Site 25, C 82
Oliver, B.C. Canada

Additional investigator(s): Mary Theberge

Objective: To determine the behavioral and ecological role of wolf howling. Also, to compare the howl characteristics of gray wolves, red wolves, and red-wolf-coyote hybrids.

Findings: Study is in progress. Howls are still being collected, and will be for some time because data collection is opportunistic rather than experimental; park rules preclude our initiation of howling responses.

Project title: Sagebrush Ecology and Ungulate Relationships

Principal investigator: Dr. Carl Wambolt
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Bozeman, MT 59717

Additional investigator(s): Scott Thompson, Harrie Sherwood

Objective: To determine 1) the current status of the sagebrush-shrub community on the northern Yellowstone mule deer winter range and 2) the importance of the sagebrush community to wintering mule deer and elk.

Findings: Mule deer utilize the several sagebrush habitat types in the boundary line area as key wintering types. They use the four woody sagebrushes and three rabbitbrushes heavily as browse, although they display a decided preference among taxa on winter range.

Project title: Ecology of Thermal Communities

Principal investigator: Dr. Richard Wiegert
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Objective: The objective of this current phase is to follow the long-term dynamics the organisms in thermal communities in the temperature range from 45 degrees C down to ambient.

Findings: The past year emphasized the extreme fluctuations in dragonfly populations from year to year. The major suspected course of this could be temperature and flow fluctuations in the stream due to variable weather. Temperature data-loggers placed directly in the stream have had minimal success due to battery failure at high temperatures and outright theft! I do have air temperatures throughout the year for almost two years, and will try to compare these to weather records from Old Faithful to see if extrapolation of the latter to Gentian Creek in other years is warranted.

Parkwide studies of the warmwater dragonfly and surveys of thermal communities are continuing. Papers are being prepared on the long-term study of dragonfly populations; the community census; and the dynamics of the alkaline thermal community that develops below 40 degrees C. In addition, I am beginning a small book on my own work and that of my students and post-docs since 1968. The title will reflect our work on the ecology of YNP thermal communities in a variety of locations.

Project title: Mycorrhizal Ecology of Thermal Sites in Yellowstone

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Objective: With this research we propose to 1) assess the distribution of mycorrhizal fungi across environmental gradients at hydrothermal sites; 2) measure the effects of mycorrhizae on plant growth in hydrothermal-influenced soils; 3) determine whether fungi in hydrothermal sites are better adapted to maximize mycorrhizal colonization benefits to host plants than fungi from non-hydrothermal sites; and 4)

increase our understanding of this plant/fungal interactions in extreme environments such as the low pH and low nutrient sites found adjacent to hydrothermal sites in Yellowstone.

Findings: Our first objective of this research was to determine whether mycorrhizae were present. We sampled plant roots from four sites including Hundred Springs Plain in the Norris Basin, Amphitheater Springs, the Firehole River near Ojo Caliente, and Rabbit Creek. We found that mycorrhizae were present at all sites, but in lowest abundance at the Ragged Hills Site adjacent to Hundred Springs Plain. At all other sites, colonization levels ranged from 6 to 54 percent, and were present in soils ranging from 3.7 to 6.5, with soil temperatures in the rhizosphere ranging from 21 to 48 degrees Centigrade. We are interested in continuing this research by investigating whether mycorrhizal fungi found at these sites are specifically adapted to site conditions, including high soil temperatures, low pH, and varying element concentrations.

ENTOMOLOGY

Project title: Butterflies of Greater Yellowstone: Aquatic Insects of Greater Yellowstone

Principal investigator: Dr. Robert Anderson
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Objective: In conjunction with two courses taught for the Yellowstone Institute at Lamar Ranch one week in early July 2000, sample butterflies and aquatic insects in the northern sector of YNP, and from Gardiner, Montana to Beartooth Summit. Specimens to be collected from meadows, fields, and ponds and small streams (catch and release only).

Findings: Butterflies representing six of the seven families of *Rhopalocera* were identified. These observations included two species of *Papilionidae* (including numerous parnassians), several species of *Pieridae, Satyridae, Nymphalidae, Lycaenidae, Hesperiidae* (sixteen species). A special effort was undertaken to identify the Yellowstone Checkerspot butterfly in the northern region, but none were found. Sex ratio information collected for parnassians indicated a predominance of males (16 males-to-4 females on the backroad to Gardiner) as noted in previous years. Samples of aquatic insects collected from various streams and glacial ponds in the northern region were similar in species composition and diversity to collections made in previous years, with approximately eight species of *Trichoptera*, several species of *Plecoptera*, about six species of *Ephemeroptera*, plus varied species of *Odonata* and aquatic *Diptera*. All specimens obtained in the park were released at point-of-capture following in-field discussions.

A special project undertaken for the Advanced Butterflies course offered through the Yellowstone Institute involved a mark/recapture study of *Parnassius phoebus*. The objective of this study was to teach the methods and value of such endeavors, using the Parnassian model and opportunity. The study was conducted from July 7 through July 22. A total of 40 *P. phoebus*, mostly males, were captured, marked/numbered and released on July 7 and 8, and from July 7 through July 22 some of these marked/numbered individuals were recaptured and released daily, some up to about 200 meters from the point of original capture. The capture/recapture information was incorporated into appropriate indices and ratios, and suggested the total size of the population of these butterflies behind the ranger station at Buffalo Ranch to be about 56 individuals on July 7, 64 individuals on July 8, and 106 individuals July 9, followed by a gradual increase to approximately 180 total butterflies when the study was terminated on July 22. Several of the numbered specimens were recaptured on more than one occasion, and the recapture data indicate these butterflies can live at least 15+ days as adults.

Project title: Butterflies of Yellowstone and Grand Teton National Parks

Principal investigator: Mr. Richard Lund
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Additional investigator(s): Mardell Moore

Objective: To produce field guides about the insects of Yellowstone and Grand Teton national parks. Photographs need to be taken of all species in the parks. Data and photos only need to be obtained. No specimens are collected. All specimens are safely netted, photographed and released live in the area of the park where discovered by the researchers.

Findings: In 2000, several more photographs of dragonflies were taken. The investigators located an area within Yellowstone where the number of different species numbered from six to eight. This observation goes counter to previous observations of, at a maximum, three different species per wetland inventoried. Investigators will be doing further research in this area in 2001. Several additional wetland areas will be inventoried in 2001. Damselfly species will be more of a focus as well in 2001.

Project title: The Mosquitos of Yellowstone National Park, a Study of Their Species and Biology

Principal investigator: Dr. Lewis Nielsen
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Holladay, UT 84117-5378

Objective: An ongoing study of the mosquito species to determine species present and their biology.

Findings: Park was not visited during 2000.

Project title: Assessment of Host Races in the Ovary-feeding Beetle, Brachypterolus pulicarius *(Coleoptera: Nitidulidae)*

Principal investigator: Dr. Robert Nowierski
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Additional investigator(s): Kelly Hering, Bryan FitzGerald

Objective: This research is being completed for Kelly Hering's Master's thesis project at Montana State University, investigating the existence of host races in the beetle, *Brachypterolus pulicarius,* a natural enemy of yellow and Dalmatian toadflax. Because this beetle exists on two separate host plants, researchers are investigating whether the species consists of two genetically distinct host races. In choosing sites to collect the insects and plant material, sites were sought where the insects have never been released as a biological control agent. Rather, they were accidentally introduced along with the weeds. Because *B. pulicarius* has never been introduced into Yellowstone, and because both yellow and *D. toadflax* infestations are present, the park offers an excellent opportunity for collecting the insects as they naturally occur on the two hosts. Along with the sites in Yellowstone, others in Canada and the northeastern United States will be analyzed.

Findings: In July 2000, Bryan FitzGerald and Kelly Hering again attempted to sample several toadflax sites. Five sites visited in 1999 were re-visited this year and efforts were made to collect insects and plant material. However, due to extremely dry conditions, no beetles were found at one of the sites. At the other four sites, a total of about 120 beetles were collected and about 10 toadflax stems were harvested. Beetles were either immediately placed into alcohol to be preserved for use in genetic analysis or were returned to campus alive to be utilized in behavioral trials prior to being preserved in alcohol. All insects collected were killed and will be processed during the course of genetic research. Behavioral trials were conducted utilizing a Y-tube olfactometer in an attempt to determine whether the beetles have a preference for the plant species they were harvested from. In 2000, no significant behavioral results were found. Genetic work has continued, and DNA extracted from B. pulicarius has been successfully amplified. Current efforts are being made to more effectively visualize Amplified Fragment Length Polymorphism (AFLP) products for genetic anlysis of intra-specific variation in the samples.

Project title: Birds, Bees, Butterflies, and Botany

Principal investigator: Mrs. Peg Steuneuberg
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Email: pegsteu@aol.com

Objective: To teach students various scientific illustration techniques for rendering insects, birds, and botanical material.

Findings: None

Project title: Respiratory Physiology and Habitat Selection in Thermophilic Aquatic Insects

Principal investigator: Dr. Brent Ybarrondo
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Objective: Understand respiratory physiology and habitat selection decisions in thermophilic aquatic insects including water scavenger beetles *(Coleoptera: Hydrophilidae)* and both adult and nymphal stages of dragonflies and damselflies *(Odonata)*. Correlate habitat selection decisions and thermal preference with respiratory physiology and development *(Odonata)*.

Findings: Adults of two species of dragonflies partition habitat riparian habitat along the Firehole River in a manner suggesting that water temperature and/or dissolved oxygen tension may be determining factors related to oviposition and/or niad development. Males of each species defend territories, in which females oviposit, that differ significantly in water temperature and dissolved oxygen. Odonate niads from thermal pools have been found to exhibit thermal preference in the laboratory, suggesting that temperature may be an important determinant of development time in these insects. Hydrophilid respiratory complex (plastron + macroplastron, or bubble) functions primarily as an oxygen reservoir at water temperatures greater than ca. 5C. Adult beetles have been collected from thermal pools during early winter and are know to overwinter as adults under thick ice in temperate ponds. Future research will investigate (1) the degree to which the respiratory complex function as a physical gill at low water temperatures (ca. Tw = 0 to 5.0C), (2) the degree to which adult male dragonflies exhibit thermal preference in controlling oviposition territories in thermally variable environment (e.g., Firehole Rive study site), and (3) development rates of odonate niads as a function of water temperature and dissolved oxygen tension will be investigated.

ENVIRONMENTAL MONITORING

Project title: A Remote Sensing and GIS-Based Model of Habitat as a Predictor of Biodiversity

Principal investigator: Dr. Diane Debinski
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Additional investigator(s): Mark Jakubauskas, Kelly Kindscher

Objective: The major objectives of the research were to 1) quantify the spatial and temporal variability in montane meadows; 2) develop a spectrally-based spatially-explicit model for predicting plant and animal (butterflies and birds) species diversity patterns in montane meadows; and 3) test the spectrally-based spatially explicit model developed in Objective 2 for predicting plant and animal species diversity patterns in montane meadows.

Findings: We sampled birds, butterflies, and plants for four years (1997-2000) in two regions of the Greater Yellowstone ecosystem: the Gallatin National Forest and northwestern portion of Yellowstone National Park and Grand Teton National Park. We used satellite imagery to classify two types of wetland meadows and four sagebrush communities. In Grand Teton National Park, our overall accuracy of mapping sagebrush communities was 65 percent, and highest for the mixed big sagebrush/low sagebrush community at 86 percent. Abundance of habitat specialist bird species was highly correlated with both meadow type and landscape variables. Butterfly species abundance and distribution was even more strongly correlated with meadow type (butterfly distribution was used to predict meadow type with a 92-96 percent accuracy in the Tetons). Voucher plants are housed at the University of Kansas McGregor Herbarium; voucher butterflies are housed at Iowa State University.

Project title: Chloride Flux Monitoring

Principal investigator: Dr. Irving Friedman
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Denver, CO 80225

Additional investigator(s): Daniel R. Norton

Objective: To provide a baseline for chloride flux exiting the park.

Findings: Chloride flux, a surrogate for heat flow, was determined for the four rivers draining Yellowstone National Park for the water years 1983 through 2000, with the exception of 1995 and 1996. The chloride that is emitted by the geothermal system underlying Yellowstone is designated "thermal chloride" and constitutes 93 percent of the total chloride exiting the park.

The Fall, Madison, Snake, and Yellowstone Rivers have been estimated to discharge 93 percent of the chloride leaving the Park, the remainder exits along the west boundary into the Henry's Fork River. The sum of the annual chloride fluxes for the four rivers varies as much as 20 percent year to year. This sum, when corrected for the climatic factors that influence this flux, shows a decline of 14 percent (0.8 percent a year) over the past 17 years. A similar decline in thermal chloride flux output from Mammoth Hot Springs has also been noted, as has a lengthening in the period of eruption of Old Faithful Geyser. We believe that these changes are related to the deflation of the Yellowstone caldera documented by changes in the ground levels surrounding Yellowstone Lake. The chloride flux for each river varied seasonally and annually and is postulated to depend primarily on the flow of hot springs. This flow, in turn, depends on the height of the local water table, which rises during spring runoff, and varies annually in synchronism with changes in precipitation.

Project title: Trace Element Content of Cervid Antlers

Principal investigator: Dr. Jack Kovach
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Objective: I am studying the strontium isotopic composition and content of strontium and other trace elements in elk and deer antlers from selected national parks in the western U.S., including Yellowstone. The study will add to the general body of knowledge about the cycling of trace elements through the environment and increase our understanding of the biogeochemistry of strontium. The study will provide baseline data from which future changes may be gauged. (A copy of the research proposal submitted to the Green Educational Foundation is on file in the Division of Research, Yellowstone Center for Resources, Mammoth Hot Springs, Yellowstone National Park. Said proposal provides a detailed description, etc. of this project.)

Findings: No significant findings to date with respect to trace-element contents inasmuch as no analytical data are yet available. Evidence of antler-chewing/osteophagia by Yellowstone elk has been obtained, and this is likely related to the major- and/or trace-element content of the antlers/bones and the nutritional status of the elk. Fieldwork by me in the park in late September and early October 2000 was directed primarily toward determining the geographic distribution and frequency of occurrence of antler-chewing/osteophagic behavior through field observations of skeletal remains of dead animals and cast (shed) elk antlers in Yellowstone's northern range.

Project title: Hydrogeomorphic Approach to the Assessment of Wetlands in YNP

Principal investigator: Mr. Chris Noble
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Additional investigator(s): Marcus Miller, Bob Leinard, Forrest Berg

Objective: Collect data in four slope wetlands in the Lamar Valley of Yellowstone National Park. This data would be added to an existing database from sites in Montana. We hope to collect data in the park for reference purposes due to lack of human disturbance and the probability that the wetlands will remain in an undisturbed condition.

Findings: We were unable to begin fieldwork this past year due budget cuts. We have received funds to continue the work in 2001.

Project title: Inventory of Wyoming Resources

Principal investigator: Mr. Dwane Van Hooser
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Ogden, UT 84401

Additional investigator(s): Michael Wilson, Bill Dunning, Dana Lambert

Objective: To collect information on the condition of forest ecosystems, estimate baseline (current) conditions and trends, and detect changes from those baselines and trends over time at the state and national level.

Findings: The Inventory of Wyoming Resources began in 1997 with installation of the ground locations. The installation was completed that same year and approximately 1/3 of the locations are re-measured each year thereafter. This project has no conclusion; therefore, there is no project ending date. As of 2000, the frequency of re-measurement and total number of locations has changed. To comply with the Farm Bill, our frequency of revising the plots is now on a five-year cycle. However, the total number of plots visited in a single year (panel) was to remain unchanged. This has resulted in our adding plots to each of the existing four panels, and in 2003, adding a complete new panel. The net increase in the design will eventually result in 2/3 more locations.

EXOTIC ANIMALS

Project title: The Invasiveness and Impact of the Exotic New Zealand Mudsnail

Principal investigator: Dr. Mark Dybdahl
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Additional investigator(s): Dr. Bob Hall

Objective: To 1) assess the invasion potential of New Zealand mudsnails by measuring their performance under ambient conditions; 2) estimate secondary production by developing a model of growth and reproduction under different ambient conditions; and 3) study the impact of mudsnails on native snails.

Findings: In the past year, my students and I have been assessing the invasion potential of mudsnails by measuring their survivorship, growth rates, and reproduction rates in both field and lab studies. In the lab this year, we raised snails from the Madison River at 12, 18, and 24 degrees C. In brief, we found that the age of first reproduction exceeds 6 months at 12 degrees. Mudsnails reproduced at 12 and 18 degrees C, but failed to produce any eggs or juveniles at 24 degrees C.

In the field, I measured growth and reproduction in three different seasons. We (Bob Hall collaborator) measured growth and reproductive rates in the Madison, Gibbon, Firehole, and Snake rivers over a two week period in July, and these measurements will be used in the model of secondary production.

I also measured growth and reproduction over a several month period at "invasion fronts" that have been identified on tributaries of the Firehole and Gibbon rivers (6 sites). These studies were conducted between September 1999 and March 2000 (3 sites), and between July 2000 and November 2000 (3 sites). I found that snails were able to grow and reproduce at sites above their current summer distributions.

The temperature regime during all of these experiments was measured using data loggers (a total of 9), which continue to monitor water temperatures. My graduate student and I established preliminary experiments to examine competition between mudsnails and native snails, and the impact of mudsnails on periphyton density. We have conducted two experiments so far (Fall 1999 and Summer 2000), both using *Physa gyrina.* Data analysis is currently underway. Samples are currently housed at Ohio University.

Project title: Linking Modeled and Experimental Interaction Strength Between Exotic New Zealand Mudsnails and Algae

Principal investigator: Dr. Robert Hall
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Address: Department of Zoology and Physiology
University of Wyoming
Laramie, WY 82071

Additional investigator(s): Mark Dybdahl, Billie Kerans

Objective: The objective is to estimate interaction strength between exotic mudsnails and periphyton by using a modeling approach and to test these estimates by using controlled field experiments. This approach will allow us to estimate impact of New Zealand mudsnails on river algae.

Findings: Using bioenergetics approaches we estimated consumption rates of algal biomass by snails in small experimental chambers in the Firehole River and Snake River in Yellowstone. We also manipulated snail densities in chambers to estimate impact to algae biomass and production over a one-week interval. In both rivers snails strongly impacted algae, as algal biomass was negatively linearly related to snail biomass. Predicted interaction strength positively related to theoretically modeled interaction strength. However, modeled interaction strength was higher than measured, likely because of high turnover of algal biomass. Growth rates of algae (i.e., production:biomass) were slower with increasing snail biomass, suggesting that snails were consuming high-turnover algal taxa. Consumption rates were negatively related to algal production rates, and algae consumption by snails consumed up to 100 percent of algae production. Given snail biomass near 25 g AFDM/m2 in the Firehole River, we suggest that these invaders are decreasing whole-stream algal production. We will reexamine these relationships this upcoming summer in the Snake and Firehole rivers.

Project title: Food Web Impacts of Exotic New Zealand Mudsnails in Yellowstone National Park

Principal investigator: Dr. Robert Hall
contact info: see above

Additional investigator(s): Mark Dybdahl, Billie Kerans

Objective: Estimate carbon flow in food webs of rivers with high (Firehole River) and low (Gibbon River) densities of exotic snails.

Findings: We are halfway through fieldwork of this study, which started field collection in August 2000. We are estimating secondary production of mudsnails and native invertebrates from two sites within the Firehole River and one site in the Gibbon River. Preliminary results show extremely high densities of snails in the Firehole River during summer and early fall (up to 400,000 / square meter), but these densities decrease during winter months. Vegetated habitat in the Firehole River has higher snail densities than riffle habitat. The Gibbon River has many fewer mudsnails than the Firehole River. We will complete field sampling September 2001 and data analyses by January 2002.

Project title: Gypsy Moth Detection Trapping

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Additional investigator(s): Tom Olliff

Objective: Detect the spread of gypsy moths.

Findings: Seventy one pheremone traps designed to attract and detect gypsy moths *(Porthetria dispar)* were placed in Yellowstone National Park in 2000. Traps were placed at all major developments, campgrounds, major attractions, and at pre-determined locations along park roads. No moths were detected in Yellowstone in 2000.

FIRE

Project title: Fire: A Force for Change and Regeneration in Natural Ecosystems

Principal investigator: Dr. John Burger
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Objective: To study the long-term changes in re-vegetation at sites affected by different fire intensities, with emphasis on lodgepole pine, aspen, and whitebark pine

Findings: Selected sites throughout the park were examined to determine differences in rate of growth of lodgepole pine seedlings/saplings and in the onset and rate of reproduction in lodgepole pine. Growth rates are minimal in some areas (10-20 cm/yr; Frying Pan Spring; Norris-Canyon site) and high in other areas (40-60 cm/yr; Madison Jct., Norris Geyser Basin; Norris-Canyon blowdown; Miller Creek trail sites). Tallest sapling measured in 2000 was 1 mi. S. of Norris Junction in an area of vigorous regrowth, 444 cm. With estimated age of 12 years. Regeneration of whitebark pine also was examined along the Hoodoo Basin trail and the Canoe Lake trail. The eastern boundary ridge between Canoe Lake and Bootjack Gap also was examined for the first time since the 1988 fires and showed signs of very heavy grazing by elk. Comparative photographs were taken at selected sites for comparison with pre-fire and early post-fire vegetation. These are/will be used in teaching modules on fire ecology and also have been used by Mary Ann Franke in her publication on research since the 1988 fires. Aspen mortality at sites near Blacktail Deer Creek appeared to be much higher in 2000 than in previous years, with about 25 percent of the above-ground stems dead.

Project title: Post-Burn Resource Selection, Physiological Condition, and Demographic Performance of Elk

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Additional investigator(s): Adam Messer

Objective: The primary objective of this research is to evaluate the consequences of the 1988 fires on elk resource selection. Selection is being quantified for populations and individuals at multiple scales ranging from selection of patches within the landscape mosaic to selection of forages and plant parts within patches. The physiological and demographic consequences of observed resource selection strategies are being assessed through noninvasive urinary and fecal assays, and telemetry. Secondary objectives include basic research on forage plant chemical compositions, plant-animal interactions and applied research to develop practical and rigorous management tools for population monitoring (aerial surveys, fecal steroid pregnancy assays, and snow-urine condition indices).

Findings: We have been successful in developing, testing, and applying a suit of research tools that is significantly enhancing our ability to address questions of animal resource selection and the physiological and demographic consequences of selection patterns. We have completed our ninth field season of data collection and maintain an instrumented population of 30-40 cow elk. Most publications to date have focused on techniques including population estimation, pregnancy assessment, and nutritional indices. This year we completed a manuscript analyzing the demographic data collected during the first seven years of research which is currently being considered by *Ecology*. Adult survival and reproduction is near the biological maximum for the species, but recruitment is highly variable, being strongly influenced by environmental variation, primarily winter severity. Despite this variable recruitment, extensive Monte Carlo simulations indicate that the population is relatively stable and is being regulated at approximately 600-800 animals. We have generated a database of greater than 9000 animal locations and are exploring a variety of analytical tools for the analysis of these data. An ArcView GIS database has been developed that integrates landscape features with all spatially-explicit databases collected on this study. We are currently developing spatially-explicit snowpack models in collaboration with NASA scientists to enhance our analyses of elk resource selection.

Project title: Dynamics of Climate, Fire and Land Use in the Greater Yellowstone Ecosystem

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Additional investigator(s): Jeremy Littell, Sean Hill, Lindsey Waggoner

Objective: To answer the following questions: 1) What was the spatial and temporal variability of pre-20th century fire? I will use these preliminary results to define the variability of fire regime as affected by elevation gradients and other topographic controls of climate. In addition, I will assess whether fire regimes have changed over the past several centuries. 2) How did climatic variability affect pre-20th century fire regimes? I will compare palecoclimatic data with pre-20th century fire data to characterize the impact of climate on fire regimes. 3) How significantly has 20th century fire exclusion changed the aboveground car-

bon pool? I will compare current aboveground carbon pools with inferred past carbon pools based on a reconstruction of the landscape. I will seek corroboration of these patterns using two model- based approaches.

Findings: In fall 2000, we selected our first field sites for this project in the northern range at Crevice Lake and Soda Butte Creek. We developed a 350-year Douglas-fir chronology at Soda Butte Creek that cross-dates reasonably well with previous sampling at Mt. Everts. We also began sampling increment cores for stand age class analysis at Soda Butte Creek. Fire scare distribution and density were assessed at both sites to apply for the initial permit. We will begin sampling these sites in spring 2001 and continue through summer 2002, fire weather permitting. Samples are archived at the Mountain Research Center.

Project title: Forest Fire Regrowth

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Additional investigator(s): Monforton School students

Objective: To collect regrowth data including percent of coverages of grasses/sedges, forbs, moss/lichens, trees, litter, bare ground. Heights of grasses and trees are also measured and recorded.

Findings: percent grasses/sedges: 14.2; percent forbs: 16.3; percent moss/lichens: 21.8; percent trees: 8.8; percent litter: 35.5; percent bare ground: 3.5; height trees: 81.9 cm; height grasses: 45.4 cm. Data from all years of the study is available at http://www.mcn.net/~monfort/YPFall2000/regrow00.html.

Project title: Impact of Fires of 1988

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Objective: Annual studies of foliage, ash and soil to track the changes in physical and chemical properties of collected materials. Sleeve tests of ash and soil. Visual microscopy of ash and soil separates. Photographic records of selected sites, measurement of ash layer, description of site.

Findings: The sites on level terrain for the most part continued to reveal an average of 0.5 inch of black

ash with small variation year by year. These sites were on forested terrain. Deviation from this result occurred on slopes that were washed by rainfall and on course soils in open terrain exposed to wind. Most dramatic is the marked increase of grasses, shrubs, and plants in the sites of level terrain in wooded areas containing considerable moisture.

Project title: Postglacial Fire Frequency and its Relation to Long-term Vegetational and Climatic Changes in Yellowstone Park

Principal investigator: Dr. Cathy Whitlock
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Objective: The primary objective has been to study the vegetational history of Yellowstone and its sensitivity to changes in climate and fire frequency. To establish a vegetational history, a network of pollen records spanning the last 14,000 years has been studied from different types of vegetation within the park. A reconstruction of past fire frequency is based on information gained from 1) a study of the depositional processes that incorporate charcoal into lake sediments; 2) a comparison of charcoal and dendrochronological records of fire occurrence during the last 750 years; and 3) an analysis of charcoal, pollen, and magnetic properties in lake-sediment cores spanning the Holocene and late-glacial periods.

Findings: Progress was made on three aspects of this project. First, the Trail Lake record has been written up and will be submitted in the coming year for publication. This record reflects the new chronology and a basal age of 8000 years old.

Analysis of the sampling of modern sediments in lakes with watersheds that were burned in 1988: this process-based study provides information necessary to interpret the charcoal record in sediment cores, by determining the time of charcoal accumulation following a fire event. This study is unique, and the results have been used by researchers around the world. The results are discussed in three manuscripts.

Third, we are collaborating with scientists from the USGS to evaluate the paleoecological history of Yellowstone lakes to past climate change. Samples have been analyzed for sediment geochemistry. Special attention has been directed to northern range lanes, particularly Crevice Lake. Crevice Lake has annually laminated sediments and cores will be taken in February 2001, as part of collaborative project with the USGS and University of Nebraska.

Other accomplishments of note are 1) publication of the fire history from Cygnet Lake in the journal *Geology*; 2) acceptance of a manuscript on the paleoecological record of plant invasions, to appear in *Western North American Naturalist;* 3) acceptance of two papers on charcoal methodology; 4) acceptance of a manuscript on the prehistory of the Rocky Mountains; and 5) presentation of results at annual meetings of the American Geophysical Union, American Association for the Advancement of Science, and Association of American Geographers.

Project title: Fire Effects Monitoring in Yellowstone National Park

Principal investigator: Yellowstone National Park Fire Effects Crew
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Additional investigator(s): Mitch Burgard, Eric Miller, Todd Carlson

Objective: Monitor the effects of prescribed fire on Yellowstone's ecosystems. Provide information to evaluate whether the objectives of the fire management office are met. Study the long-term effects of fire on the landscape.

Findings: The Yellowstone Fire Effects Crew established several plots ahead of naturally occurring wildfires during the particularly active 2000 fire season. We installed five monitoring plots ahead of three fires: Two-Smokes, Boundary and Plateau. The first plot (Two-Smokes) was installed in a smoldering tree island in the meadow complex on the Pitchstone Plateau in the southwestern corner of the park. In the following weeks the fire burned the rest of the tree island and spotted across the intervening meadow into two other tree islands. The plot was resampled on September 13.

We installed two more plots on the Boundary Fire at the south end of the park. This fire was ignited by lightning on August 15 in a forest of twelve year old lodgepole pines regenerating an area that was overrun by crown fire in 1988. One plot burned on August 22, a day after installation. The other burned about five days later. These two plots are interesting because the young cover type (LP0) was not expected to carry fire as well as it did. Fire carried through the sedges when possible and otherwise resided in rotten logs outside the burn period. We found that old, rotten logs were completely consumed while the more solid logs resulting from trees killed in 1988 were only scorched and partially burned. We found that the fire would not consume logs with a bulk density (a measure of "rotten-ness") greater than 0.34 grams/cm3. This figure, as well as the vegetation data from this year's sampling and future resampling, may be applied to the vast areas of the park that are regenerating from the fires of 1988 and will aid our predictions of future wildfire behavior in this cover type. The last two plots were installed on the Plateau Fire near Buffalo Lake Cabin but neither of them burned.

We also visited two fire plots established in 1977 and 1988 by Don Despain. Despain's dataset is comprised of twelve plots installed between 1977 and 1989 and periodically resampled every few years thereafter. We presented some of these results at the Second USGS Wildland Fire Workshop in Los Alamos in early November, and are currently working toward publishing this information in a paper. We installed one plot in the proposed prescribed burn unit at Grant Village. This plot, in addition to several other plots in the burn unit will allow us to ensure fire management's objectives are met, and to monitor post-fire effects resulting from the burn.

FISHERIES MANAGEMENT

Project title: LeHardy's Rapids Yellowstone Cutthroat Egg Collection for the Development of Species-specific Broodstock for Drainage Restoration

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Additional investigator(s): Joe Gillis, Steve Sharon, Dave Miller, Paul Kretschmar

Objective: To collect and fertilize eggs for Yellowstone cutthroat trout pairs to develop a captured broodstock program. Eggs will be collected from the population that inhabits the Yellowstone Lake to the upper falls. Fish management at Yellowstone has also asked for egg collection on various other tributaries to aid in whirling disease research. The primary capture location will be LeHardy's Rapids, although other sites within the drainage may be considered if catch rates do not meet objectives. The original objective each year was to collect a partial spawn from a minimum of 25 pairs for four consecutive years (1993-1996) for the purpose of stock recruitment of a broodstock to be held at Clark's Fork Fish Hatchery. This broodstock will be used for drainage restoration of the endemic range of the Yellowstone River in Wyoming and also assist in the restoration projects in Montana.

Findings: We have had excellent cooperation with YNP personnel in accomplishing our goals with this project. This is the last anticipated year of the consecutive year collection process. Year 2000 collection will assure the broodstock adequate year classes for its development. We will then return to YNP every 3–5 years to infuse new genetic material from the LeHardy's rapids stock into our Clark's Fork Fish Hatchery Stock to assure good genetic representation of this stock.

Project title: Collection of Gametes from Wildstock Lewis Lake Lake Trout to Establish a Captive Broodstock to Support Lake Trout Restoration in the Great Lakes

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Additional investigator(s): Ed Stege

Objective: The objective of this research project was to capture and spawn a minimum of 50 pairs of Lewis

Lake lake trout in calendar year 2000. Fertilized eggs were to be transported to the Saratoga National Fish Hatchery, Wyoming, for the establishment of a captive broodstock. Progeny from the Lewis Lake captive broodstock at Saratoga NFH will then be used for lake trout restoration/recovery programs in the Great Lakes. This project is part of an ongoing effort that was initiated in 1983 with respect to utilizing Lewis Lake lake trout for lake trout restoration in the Great Lakes. Genetic considerations mandate an infusion of wild genes into captive broodstocks at regular intervals to insure the genetic integrity of broodstock populations.

Findings: Adult lake trout were captured with the assistance of Yellowstone National Park personnel using gillnets set at near-shore spawning locations. A total of 68 paired matings were effected. Eggs from five females were split and fertilized individually with milt from different males, resulting in a total of 85 genetically distinct egg lots. Upon completion of spawning, all lake trout were returned to Lewis Lake. All egg lots were transported to the Saratoga NFH and held in quarantine until completion of a complete disease evaluation.

Project title: Cutthroat Trout Egg and Sperm Collection

Principal investigator: Mr. Daryl Hodges
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Additional investigator(s): Montana Fish, Wildlife and Parks personnel

Objective: To succesfully manage Montana's fishery resources, we need to maintain our hatchery broodstocks with a wide genetic diversity. These broodstocks should mirror their wild ancestors as closely as possible. The original gametes for our Yellowstone cutthroat trout Broodstock came from McBride Lake in Yellowstone National Park in 1969. The last time gametes were taken from the lake to supplement the broodstock was 1987. To once again infuse our broodstock with new genetic material, we will collect gametes from Yellowstone cutthroat trout in McBride Lake for three consecutive years beginning in 2000. We will take gametes from 10 pair of fish each year. Health and genetic samples will be taken from the fish gametes are taken from.

Findings: Due to the failure to get approval for lethally sampling 60 fish for genetic testing and disease certification, eggs were not collected in 2000. FWP Fisheries Biologists, hatchery employees and Fish Health Biologists are prepared to go to McBride Lake in the spring of 2001 and the following two years if the needed approval is received.

Project title: The Spatial and Temporal Spawning Distributions of Yellowstone Cutthroat and Rainbow Trout in the Upper Yellowstone River Drainage

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Additional investigator(s): Bradley Shepard, James De Rito

Objective: This research project will radio-track spawning Yellowstone cutthroat trout (YCT), rainbow trout, and hybrids of the two species to determine where and when these three groups spawn.

Findings: Radio-tags were implanted into 50 fish during April 4 and 5, 2001. Fish are being relocated on a weekly basis. Some rainbow trout and hybrids have moved into tributaries (outside park) to spawn. Fish that were tagged in the Corwin Springs, Montana area of the Yellowstone River may move upriver or into tributaries that are accessible in Yellowstone Park.

FORESTRY

Project title: Remote Sensing-based Geostatistical Modeling for Coniferous Forest Inventory and Characterization

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Additional investigator(s): Edward A. Martinko, Kevin P. Price

Objective: The goal of this research is to develop, test, and demonstrate an integrated remote sensing and geostatistical approach for the analysis of forest canopy structure, secondary forest regrowth, and forest fire history that takes advantage of the spectral and spatial correlation of ground phenomena and remotely sensed information. The project has four objectives: 1) development of geostatistical models for forest biophysical parameters (height, density, basal area, leaf area index, and biomass) using multiscale satellite imagery and field data; 2) calibration and verification of the models by field data and statistical means; 3) testing the models in two specific forest characterization and inventory applications, (forest cover type mapping and insect damage assessment); and 4) dissemination of the algorithms and procedures to the user community via online tutorials and software modules. Initial model development will focus on the lodgepole pine forest of the Greater Yellowstone Ecosystem.

Findings: Key milestones achieved: 1) additional summer 2000 field campaign in Yellowstone National Park: 122 sites sampled for forest biophysical characteristics in Central Plateau; 2) LANDSAT 7 Thematic Mapper data of Yellowstone for July, August, September 2000 acquired from EROS Data Center and processed; 3) project web page developed and is online at http://www.kars.ukans.edu/forest. Planned work for summer 2001 includes additional forest sampling and measurement of forest spectral reflectance characteristics in the Central Plateau coincident with a special satellite data acquisition by NASA EO-1/Hyperion satellite, and field checking of model predictions of forest parameters. Preliminary predictive maps of Yellowstone forest density, height, basal area will be created using field and satellite image data. Analysis in 2001-02 will produce predictive maps of forest fire danger, insect infestation assessment, and forest cover types. Yellowstone Area Online Image Data archive will be established to provide LANDSAT 7 satellite imagery to any interested researchers at http://www.kars.ukans.edu/forest/landsat7.html.

FUNGI

Project title: Fungi from Geothermal Soils and Thermotolerant Plants

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Additional investigator(s): Dr. Regina Redman, Dr. Joan Henson, Ms. Kathy Sheehan

Objective: To provide information to increase our understanding of fungal survival in unique environments, the roles of fungi in ecosystem dynamics, and the temporal and spatial scales of the micro-habitats that fungi occupy. Specifically, this work will provide information about 1) how fungi survive under environmental conditions too harsh for mycelial growth; 2) whether fungal community structure changes in response to environmental conditions; 3) if fungi can alter between saprophytic and symbiotic lifestyles in response to environmental conditions; 4) the scale of soil studies necessary to accurately assess the roles of these fungi in ecosystem dynamics; 5) how biological and/or genetic diversity of fungal communities changes in response to environmental conditions; 6) the adaptive mechanisms of tolerance required for the growth of fungi soils containing high levels of metals and other inorganic chemicals. In addition, the feasibility of developing molecular biological tools will be determined for rapidly assessing a) fungal community structure based on molecular biomass measurements; b) the metabolically active, and inactive, species of fungal communities; and c) the occurrence of fungi in thermotolerant plants.

Findings: Several fungal species have been isolated from geothermal soils and found to be either mesophilic or thermophilic. The populations of both fluctuate throughout the year as a result of soil temperature and moisture. Fungi are in highest densities in soil under plants and can be found in soils with temperatures up to 100 C. The annual temperatures of the geothermal soils fluctuated as much as 30 - 40 C while non-thermal soils fluctuated 5 - 10 C. There are two classes of fungi present in the soils: saprophytic and symbiotic. The symbiotic fungi colonize the dominant plant species in the geothermal soils *(Dichanthelium lanuginosum).* Preliminary data suggest that the fungal symbiont may be responsible for the ability of the plant to tolerate high temperatures, dry summers, and heavy metals. This project is still in progress.

GEOCHEMISTRY

Project title: Oxygen Isotope Geochemistry and U-Pb Dating of Zircons of Yellowstone Volcanic Rocks

Principal investigator: Dr. Ilya Bindeman
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Additional investigator(s): John Valley

Objective: To 1) understand the generation of oxygen-18 depleted rhyolites inside of Yellowstone caldera; 2) estimate zircon crystallization and residence time in silicic magma chambers; and 3) date volcanic units using U-Pb analyses of zircon with an ion microprobe.

Findings: Oxygen-18 depleted rhyolites were produced as a result of total remelting of hydrothermally altered rocks in the downdropped caldera block. Two papers are written (Bindeman and Valley, "Formation of low-delta-18-O rhyolites after caldera collapse at Yellowstone," *Geology* 28, n8, p. 719-722.; and Bindeman and Valley (2000) "Low-delta-18-O rhyolites from Yellowstone: Magmatic evolution based on analyses of zircons and individual phenocrysts." *J of Petrology,* August 2001). Dating of zircons with an ion microprobe revealed that the majority of zircons in post-Lava Creek tuff, and post-Huckleberry Ridge tuff intracaldera lavas is inherited from the older Yellowstone rhyolites. A paper (Bindeman, Valley, Wooden, Persing (2001) "Post-caldera volcanism: In situ measurement of U-Pb age and oxygen isotope ratio in Pleistocene zircons from Yellowstone caldera," *Earth and Planetary Science Letters*,) is accepted for publcation. U-Pb zircon ages of Lava Creek Tuff and Huckleberry Ridge Tuffs are similar to Ar/Ar ages.

Project title: Investigation of CO2 Emissions Related to the Yellowstone Volcanic/Hydrothermal System

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Additional investigator(s): Cindy Werner

Objective: To 1) estimate the CO2 emissions from the Yellowstone volcanic/hydrothermal system; 2) monitor background temporal variability of CO2 emissions, and how variations are related to changes in hydothermal and seismic activity; 3) study the spatial distribution of CO2 emissions and investigate controls on spatial heterogeneity of gas emissions; and 4) monitor gas chemistry including carbon and helium

isotopes to gain a broader understanding of the sources of gas emissions and interactions with the hydrothermal system.

Findings: A stratified-adaptive sampling plan was designed to estimate CO2 degassing in Yellowstone National Park, and applied in the Mud Volcano thermal area. The stratified component focused effort in regions with the most spatial heterogeneity (high-flux regions). The maximum and minimum measurements for vent and diffuse fluxes were 1.7 x 10^8 and 6.3 x10^4 mols/yr, and 32,000 and 4.0 g/m2day, respectively. Fluxes observed in most vegetated regions of Mud Volcano were similar to values reported by agricultural studies (less than 38 g CO2/m2day). However, we also found a few high-flux vegetated sites (up to 5,000 g/m2day) that are likely thermal features that have waned in thermal activity, yet are preferred pathways for degassing of deep CO2. Vent degassing (0.76 to 2.5 x 10^9 mols/yr) accounts for 32 to 63 percent of the total degassing observed at Mud Volcano (2.4 to 4 x 10^9 mols/yr). Temporal variation of CO2 emissions was observed to correlate with soil moisture, and environmental conditions. Results were published in Werner, et al, 2000, "CO2 Emissions related to the Yellowstone Volcanic System: 2: Statistical Sampling, Total Degassing, and Transport Mechanisms," *Journal of Geophysical Research*, Vol.105, B5, 10,831-10,846 and Boomer, K., Werner C., and Brantley, S.L., 2000, "CO2 Emissions related to the Yellowstone Volcanic System: 1: Development of a Stratified Adaptive Cluster Sampling Plan," *Journal of Geophysical Research*, 105, B5, 10,817 -10,830.

Surface degassing measurements in the Obsidian Pool region of Mud Volcano were used to test eddy correlation (a micrometeorological technique) as a means to measure volcanic and geothermal emissions. Results of this preliminary investigation suggest that eddy correlation is a viable alternative to typically used chamber methods used to measure degassing in geothermal areas. Results were published in Werner et al., 2000, "Eddy-Correlation Measurement of Hydrothermal Gases," *Geophysical Research Letters*, Vol. 27, No. 18 , 2925-2929.

Preliminary investigation of the CO2 emissions in Upper Geyser Basin; Mammoth Springs; Roaring Mountain; Washburn Springs; Crater Hills; and the Lamar River Valley suggest that diffuse degassing is highest in acid-sulfate and travertine precipitating regions, and lowest in regions of silica precipitation and sulfur flows. Further degassing measurements are planned for summer 2001. We are currently estimating the CO2 emissions for the Yellowstone system using geologic and geochemical constraints.

Project title: Water Chemistry and its Relationship to Local Geology: A Yellowstone Case Study

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Additional investigator(s): Clara Cotten

Objective: This study is an ongoing component of Geology 329 taught from the Indiana University

Geologic Field Station, Cardwell, MT. During the course of the field-based class, undergraduate students involved with several environmentally oriented programs on campus are involved in their first major field experience. The objectives of this study are two-fold. First, the Yellowstone field trip provides a unique opportunity to look at an ecosystem that is heavily influenced by hydrothermal activity, which is in stark contrast to the riparian and montane systems in the course study areas of the Tobacco Root Mountains. During the weeks preceding the Yellowstone trip, the students engage in the collection of field measurements of various aquatic systems encountered in their study areas. This data set (including oxidation-reduction potential, pH, temperature, and specific conductance), us used as a comparative set against the data collected in the thermal features of Yellowstone.

Findings: The findings of the 2000 field season indicate that within the Norris Geyser basin, there have been remarkable shifts in the heat centers during the last 40 years. There are several springs that are no longer active at the surface, and new springs have developed. The centers of highest recorded temperatures have shifted within the basin and are typically to the west of the major 1960s centers of highest temperatures. Again, this was an excellent illustration of the ephemeral nature of these features.

The visit to the backcountry spring from the Firehole Lake Drive was an excellent illustration of the influence of temperature on the other parameters of aqueous geochemistry. Careful measurements of the oxidation-reduction potential, pH, and specific conductance were analyzed and the values plotted with distance from the main pool. The chemical parameters recorded were well matched to the physical features observed (microbial community color, precipitation of mineral phases, and adjacent vegetation).

The overall Yellowstone experience of the course participants was one of the most highly rated portions of the course. Within a very concentrated area, students were able to observe and apply principles of aqueous geochemistry and in turn observe the influence the waters have on the communities of plants and animals around them. We hope to extend the research permit to continue the development of the course database for the students work with.

Project title: Geochemistry and Geochronology of Eocene Potassic Volcanism in the Absaroka Volcanic Field

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Additional investigator(s): Charles Lindsay

Objective: Our objective is to carry out a geological and geochemical transect across the northern part of the Eocene Absaroka volcanic field. Because the Absaroka volcanic rocks record one of the most voluminous and compositionally diverse magmatic episodes to affect the Cordillera during the Eocene, the results obtained from this study will improve our knowledge of the ages, compositions, and petrogenesis of Tertiary magmatism in the northern Rocky Mountains. This, in turn, will provide insight into the funda-

mental problem of how rock suites with arc-like geochemical features can form in such different tectonic environments and possibly in the absence of contemporaneous subduction. The targeted areas in Yellowstone National Park are the Mt. Washburn-Observation Peak volcanic center, the Sepulcher Mountain-Electric Peak eruptive center, and the Sylvan Pass-Eagle Peak eruptive center.

Findings: Our work in the Absaroka volcanic rocks during the previous year is summarized as follows: Whole-rock major and trace element abundances were determined for 40 stratigraphically constrained lava flows from Sepulcher Mountain (SM) and 17 hypabyssal intrusive units at adjacent Electric Peak (EP). SM lavas are predominately augite +/- enstatite basaltic-andesites, hornblende andesites, and hornblende +/- biotite dacites. EP intrusive units include biotite +/- hornblende quartz diorites, tonalites, and granodiorites. Petrographically, both intrusive and extrusive samples exhibit strong evidence of mixing and/or assimilation (mafic inclusions, resorbed cellular plagioclase, heterogeneous glass/groundmass compositions, and partially resorbed xenoliths). Rocks from SM and EP are compositionally similar and define a high-K, calc-alkaline series with compositions ranging continuously from 55-73 wt percent SiO2, 1.9-4.0 wt percent K2O, and Mgno. from 56-27. Major-element variation diagrams define roughly linear arrays, while the compatible trace elements Ni and Cr define both mixing and fractionation trends. Trace element abundances are characterized by extreme LILE (Ba, Rb, K) enrichment (100-400 X chondrite, ~2000 ppm Ba) and HFSE (Nb, Ta, Zr, Hf) depletion (3-50 X chondrite). Chondrite-normalized REE patterns are broadly similar for all rock types, characterized by concave-up fanning patterns, strong LREE enrichments (Ce/Yb n = 10-36), and no Eu anomalies. Some evolved magmas have lower REE abundances than mafic magmas. The large range in incompatible trace element ratios (Hf/Ta = 7-12 for the least evolved magmas and K/Rb = 275-575 for the entire suite) are consistent with REE abundances and suggest that the suite cannot be related to a single parental magma by simple fractionation processes. Based on these data, we hypothesize that the calc-alkaline signature results from mixing with and assimilation of crustal melts (coupled with fractional crystallization) by multiple parental compositions. The calc-alkaline nature of the magmas appears to be inherited from interaction with continental crust.

Project title: Biogeochemistry of Hydrothermal Springs

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Additional investigator(s): Cindy Wilson, William Cooper

Objective: 1) Investigate geochemical variations in microbial mats, pore waters, siliceous sinters and geyserites at different hot springs and thermal drainages in the park. Results will be compared with stages of silica diagenesis and geochemistry; 2) Investigate the local hydrogeological characteristics of hot spring-influenced drainages. Results will be used to calculate mass balances for such drainages, to determine silica

deposition rates in sinter mounds and to determine interaction with local groundwater; 3) Investigate photochemical processes in thermal springs of various composition.

Findings: During the past year, work focused on expanding research activities in the area of hydrogen peroxide cycling. The topic has become far more significant than previously realized. Originally of significance as an environmental factor, it is now clear that the presence of reactive oxygen species, of which hydrogen peroxide is one, is significant in determining internal and external responses of organisms to stress. Specifically, it has implications for enzyme evolution and for cellular function and repair. Implications for the conversion of solar energy to chemical energy are of equal importance. All of these topics are relevant to efforts to assist in the design of better tests for life on other planets as well as in understanding the origin of life on our own planet.

Much of the effort on the geochemistry of hydrothermal springs in 2000 focused on hydrogen peroxide production. Hydrogen peroxide production and cycling was essentially absent in all thermal springs examined likely as a consequence of fire-induced haze. The haze filtered nearly all of the UV radiation, reducing photochemical reactions to extremely low rates. The decay rates, as measured on field samples, were higher than values previously measured. As a consequence, hydrogen peroxide concentrations were not observed at detectable levels. Other biogeochemical cycles were also suppressed; iron and sulfur cycling was not observed.

Project title: Arsenic Biogeochemisry in Yellowstone National Park

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Additional investigator(s): Timothy McDermott, Colin Jackson, Heiko Langner

Objective: Our work will focus on geochemical and microbiological processes that influence the speciation and behavior of arsenic in thermal environments. Given the toxicity and potential negative impacts that arsenic (As) may have on biota in non-park environments, the thermal springs represent a potentially informative model system to begin to understand how microbiological life forms metabolize or detoxify As.

Findings: We obtained data on the oxidation of arsenite to arsenate in several hot springs in the Norris Geyser Basin. A detailed study was conducted in one acidic spring where rates of arsenite oxidation were determined along with a suite of other aqueous and sediment characteristics. Arsenate oxidation seems to be linked to the disappearance of sulfide from the spring water and to the presence of certain thermophilic microbial species.

Project title: Sulfur Speciation and Redox Processes in Mineral Springs and their Drainages

Principal investigator: Dr. D. Kirk Nordstrom
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Additional investigator(s): Gordon Southam

Objective: The primary objective is to determine the actual speciation of dissolved sulfur species as they undergo oxidation and volatile losses of H2S. Intermediate sulfoxyanions such as thiosulfate have been implicated as complexing agents to solubilize and mobilize metals in the formation of ore deposits and as monitors of volcanic activity. We hope to relate sulfur speciation in hot springs and their overflow drainages to rates of oxygen diffusion and solubility.

Findings: We have found thiosulfate in many hot springs throughout the park and we find it primarily formed from the oxidation of H2S. In a few rare cases the concentration of thiosulfate on a molar basis (or weight basis) is present in higher concentration than H2S. For these springs, thiosulfate is the main form of reduced sulfur dissolved in the water. These data and their interpretations can be found in 2 USGS open-file reports and in 2 scientific publications listed in the bibliography. One pool in particular, Cinder Pool, not only has high thiosulfate but is the only place where polythionate has been found. In this pool thiosulfate is likely to be forming from hydrolysis of a molten sulfur layer known to exist at 18-20 meters depth. The polythionate has been shown to be formed from thiosulfate and catalyzed by pyrite that occurs within the floating cinders on the pool's surface. One scientific paper is devoted to a description of the complex chemistry of sulfur at Cinder Pool. Any evidence for thiosulfate to be important in the formation of hydrothermal ore deposits is largely discredited.

Project title: Honors Freshman Quantitative Analysis

Principal investigator: Dr. Christopher Parr
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Objective: To give honors freshman chemistry students (CHM 1215) a real-world example of a total alkalinity titration.

Findings: Total alkalinity titrations were performed as described in Harris's Quantitative Chemical Analysis, but the many months intervening between collection and analysis rendered the results useful only

as an exercise.

Project title: Field Trip to Yellowstone National Park, Water Sampling

Principal investigator: Dr. Jeffrey Rosentreter
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Objective: Investigation of water composition at a variety of park locations in an effort to correlate the original fate and history of the solutions.

Findings: Several years of collection trips have identified a variety of geothermal surface features including neutral chloride, acid sulfate, and alkaline carbonate. Additionally, surface waters in the Sheepeater Cliff area have to some degree been linked top the spring waters at Mammoth Hot Springs. Surface waters near Madison Junction were thought at one time by our researchers to be contaminated with municipal waste treatment contaminants, but this past year we determined that excessive chloride detections were from natural sources.

Project title: Geochemical and Geophysical Investigations of Mine Impacts and Watersheds, Yellowstone National Park

Principal investigator: Dr. Ed Schrader
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Additional investigator(s): Dr. James Harris, Lori Eversull, Dr. Allen Bishop

Objective: The fundamental goals of this project are to contribute to the understanding of the complex geochemistry of the Soda Butte Creek watershed, and to investigate the impact of mining activities near the creek's headwaters. This is accomplished through: 1) building a long-term database documenting seasonal variations in stream chemistry and metal concentrations in stream waters and sediments, and 2) delineation of shallow subsurface features in the Soda Butte Creek floodplain.

Findings: Elevated concentrations of metals (Cu, Zn, Fe, Ag) are recorded in sediments immediately downstream of the McLaren mine tailings. Concentrations decrease sharply with distance from the tail-

ings, and generally return to background levels within four miles downstream. Although the mine tailings are a significant source of contaminants, Republic Creek also contributes substantial amounts of nickel and related metals; nickel concentrations remain elevated throughout the length of Soda Butte Creek to the Lamar confluence. Hydrothermal tributaries at Soda Butte Mound and Warm Creek influence stream chemistry, at times contributing significant dissolved species. Seasonal variations in contaminants are evident. Low flow conditions generally favor higher concentrations in sediments, while high flow conditions tend to dilute concentrations and distribute contaminants a greater distance downstream from the source. This study will continue with field measurements and sampling of stream sediment and waters on a seasonal basis.

Project title: Geology and Geochemistry of Thermal Springs in YNP

Principal investigator: Dr. David Wenner
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Objective: To teach students the interrelationship between the geological setting and geochemistry of thermal springs.

Findings: The Department of Geology at the University of Georgia conducted an eight week summer field course in geology and anthropology in 1999. As part of this course, a group of 24 students, this instructor, and 3 teaching assistants visited various thermal areas within YNP on July 30, 2000.

GEOGRAPHY

Project title: Spatial Patterns and Correlations to Socioeconomic Factors of Environmental Impact Statement (EIS) Public Comments: a Yellowstone National Park case study

Principal investigator: Mr. Eric Compas
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Objective: Spatial analysis of Environmental Impact Statement (EIS) public comments is traditionally limited to a tally of how many responses were received from each state or country. More in-depth analysis of the spatial component of public comments and the correlation of these comments to other spatial data sets may lead to new information about these comments and the individuals submitting them. This research will attempt to look for spatial patterns in the submitted comments and correlate these patterns to socioeconomic variables such as urban/rural residence, income, and education level.

The results of this study may give a better understanding of the spatial origins of specific comments and the socioeconomic backgrounds of comment givers. Which socioeconomic groups' views are represented in public comments? Which are not? If there are spatial patterns to various management scenarios, what are they? Finally, this study will better determine the usability of public comments to answer questions such as these.

Management implications include targeting public statements and forums to broaden the reach of EIS documents; improving the solicitation of public comments from socioeconomic groups that are not represented; and learning more about where support or opposition for specific alternatives is originating.

Findings: none to date

Project title: High School Physical Geography Field Study: Evaluate the Correlation Between Temperature, Slope, Fluvial Action and the Development and Characteristics of Thermus Aquaticus

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Objective: Students conduct field testing and observations at Narrow Gauge in Mammoth. Class projects vary.

Findings: Projects vary for each group.

GEOLOGY

Project title: The Search for Microbial Biomarkers in Terrestrial Deposits

Principal investigator: Dr. Henry Chafetz
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Additional investigator(s): Sean Guidry

Objective: The primary purpose of this investigation is to evaluate the fossilization process and the potential for a long term record of the microbial life that exists associated with hot springs and their carbonate (travertine) and siliceous (siliceous sinter) deposits. Basically, we are looking for biomarkers (indicators that microbes once existed as part of the hot spring environment). This will allow us to determine the likelihood of finding fossilized microbes in extraterrestrial bodies, e.g., Mars, and what is the most likely preserved material. For example, will we have a better chance of finding body fossils or geochemical indicators of former organisms.

In order to carry out this investigation, we are analyzing the waters from which the mineral precipitates originate as well as the solid precipitates of carbonate (Mammoth Hot Springs) and siliceous sinter (Cistern Spring, Norris Geyser Basin). It is our intent to search for mineralogical (crystal habit, size, etc.) and geochemical (major, minor, and trace elements as well as isotopic) differences between biotically induced and abiotic precipitates as well as microbial remains (bacterial body fossils, biofilms, etc.).

Findings: Our work to date has indicated that biogenic compounds appear to break down relatively quickly in this hot environment and thus will not be well preserved in the ancient record. Some body fossil types, however, seem to display relatively good preservation potential.

Project title: Volcanology and Petrology of the Yellowstone Plateau Volcanic Field

Principal investigator: Dr. Robert L. Christiansen
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Additional investigator(s): Wes Hildreth

Objective: To understand the origins and eruptive mechanisms of late Cenozoic volcanic activity in the

region of Yellowstone National Park and complete systematic geologic-mapping studies carried out intermittently in the park region since the 1960s.

Findings: No new work was done on this project in 2000.

Project title: Aqueous-Solid Geochemical Process Model of Travertine Precipitation at Angel Terrace, Mammoth Hot Springs, Yellowstone National Park

Principal investigator: Dr. Bruce Fouke
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Objective: This research will develop a quantitative process model that will incrementally track the diagenesis of hot spring travertine depositional facies from the modern through the Recent, Holocene, and Pleistocene. Resulting constraints on the rates and products of water-rock reactions provide the template necessary to accurately interpret travertine deposits in the early Earth and perhaps other planets. Travertines form where carbonate minerals precipitate near the vents of terrestrial hot springs, thus housing important information on water chemistry, hydrologic transport, climate, and microbial assemblages. There are, however, no systematic studies that offer guidance on how to interpret the complex crystalline fabrics and chemistries inherent to fossilized hot spring travertine. The model developed in this proposal will considerably improve our ability to extract original environmental information from ancient travertines by directly linking crystalline fabric and chemistry with original aqueous and post-depositional diagenetic processes.

Findings: The rate of crystal formation in each facies is dependent on CO2 degassing, temperature, pH, and microbial interactions. Crystal growth rates and patterns from two different pond facies have been measured in detail. Pond travertine varies in composition from aragonite needle shrubs ('fuzzy dumbbells') to rigid networks of ca1cite and aragonite. The first pond is located roughly 6 meters from the main vent along a secondary flow path. The temperature of this pond water varies from 39*C at the entrance to 37*C at the lip of the pond, while the pH ranges from 7.985 at the entrance to 8.092 at the pond lip. Isotope analysis of the pond water shows distinct positional variation, with dC13/dO18 values of (2.570, -16.720), (2.870, -16.720), and (2.340, -16.690), corresponding to the entrance, the center, and the lip of the pond. The travertine deposited showed significant variation with location within the pond. Average travertine precipitation was measured at 80 mm/day at the entrance, 50 mm/day at the center, and 125mm/day at the lip of the pond. The porosity of the deposits increased dramatically at the edges of the pond. The second pond, located roughly 2 meters away from the secondary vents along the main flow path, exhibits significantly different water chemistry and travertine deposition. In this second pond, the temperature varied from 60.9*C at the entrance to 58.5 *C at the lip, while the pH ranged from 7.320 to 7.385 over the same range. Travertine deposition at the lip of this pond was over 400 mm/day and con-

tained a large degree of porosity. From the comparison of these two ponds, it is apparent that crystal growth is strongly correlated to water flow, temperature, and pH. From analyzing the crystal growth structures of travertine, it is possible to draw conclusions about the water chemistry at the time of deposition. With this understanding, it is possible to compare travertine deposition to other carbonate deposits, such as those contained within ALH-84001.

Project title: Mapping the Mineralogy, Vegetation and Microbiota of Yellowstone National Park Using Imaging Spectroscopy

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Additional investigator(s): Eric Livo, Roger Clark

Objective: We are applying advanced remote sensing methods to map the distributions of vegetation (forest and nonforest species). Furthermore, we are applying these data to the mapping of microbial mats in the hydrothermal systems of Yellowstone, including the Upper and Lower Geyser Basins, Norris Geyser Basin, and Mammoth Hot Springs. In addition, the mineralogy of exposed rocks and soils on the surface are being characterized with this remote sensing data.

Findings: The results have successfully demonstrated the imaging spectroscopy using AVIRIS data can discriminate between the conifer cover types of Yellowstone. Specifically, the occurrences of whitebark pine, lodgepole pine, Douglas-fir, and a mixed Engelmann spruce/subalpine fir forest types were mapped in the park. The results showed that high altitude surveys by airborne spectrometers with 17 meter pixel size were able to distinguish microbial reflectance signatures from other materials. These unique signatures were used to map the occurrence of hot springs for selected flight lines over the park. Mineralogy of the hydrothermals systems, mapped by AVIRIS and the USGS Tetracorder system, revealed the different types of mineral alteration of bedrock in the park. For example, occurrences of carbonates, clays, alunite and siliceous sinter were identified.

Project title: Quaternary Geology and Ecology of the Greater Yellowstone Area

Principal investigator: Dr. Kenneth Pierce
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Additional investigator(s): Don Despain, Jon Wraith, Cliff Montagne, John Good

Objective: To determine 1) the history of glaciation and distribution of glacial deposits; 2) the history of Yellowstone Lake and River level changes and related history of caldera inflation and deflation episodes; 3) the history of hydrothermal explosions, particularly in the Yellowstone Lake area; 4) geologic controls of plant ecology in the Greater Yellowstone area (with Don Despain and Ann Rodman); 5) the history of climate and fires, particularly by coring Crevice Lake; 6) the history of glacial and other floods, particularly along the Yellowstone River; 7) the history of faulting, particularly in the Mt Sheridan area, West Yellowstone Basin, and the upper Yellowstone Valley; and 8) to assist Interpretive staff in presentation of geologic topics to the public; and 9) evaluate the concept of the Yellowstone hot spot.

Findings: Nearly all of Yellowstone was glaciated during the last glaciation. During glacial buildup and recession, glaciers advanced from the surounding mountains onto the Yellowstone Plateau, whereas during full-glacial conditions, a large ice cap built up on the Yellowstone Plateau. Plant ecology is strongly controlled by geology, including lodgepole pine forests on the sandy, nutrient-poor rhyolite soils and areas of fine-grained glacial deposits largely in grasslands. Yellowstone Lake was below present levels about 3,000 years BP, and major rises in lake level occurred between 2,800 yr BP and present and between about 8,500 and 7,500 years BP.

Project title: Study and Monitoring of Selected Geyser Activity

Principal investigator: Mr. Ralph Taylor
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Objective: To study the activity of selected geysers in the Upper Geyser Basin, Lower Geyser Basin, and West Thumb Geyser Basin to determine the activity patterns during the study period. This information will provide baseline activity data for these geysers. A long-term record of geyser activity will be compared to other factors such as precipitation, lake level (for geysers near Yellowstone Lake) and activity of nearby geysers to determine possible influences of these factors on geyser activity.

Findings: During summer and fall 2000, nine electronic data loggers were deployed on geysers that have been monitored in previous years as part of this study and on some additional geysers. Data for all of the geysers was recorded, analyzed, and summary statistics and graphs of the activity for the monitoring period (June 24, 2000 through October 6, 2000) were prepared. Several of the loggers are deployed for the winter of 2000-2001. In a few cases, equipment failures caused loss of data. The following describes the geysers monitored and the years for which data are available: Aurum Geyser (1997-2000), Boardwalk Geyser (1998, 2000), Depression Geyser (1997-2000), Lion Geyser (1998-2000), Little Cub Geyser (1998-

2000), Plate Geyser (1998-2000), Pyramid Geyser (1995-2000), Lone Pine Geyser (1997-2000).

Data from 2000 indicates that Plate Geyser and Boardwalk Geyser are interconnected. Several Geyser Hill geysers show long-term (~5-7 days) variation in intervals. Intervals between eruptions of Lone Pine Geyser at West Thumb show some variation that may be related to lake level.

Software to extract eruption data from the temperature record was improved and extended this summer. All data files are archived on the Principal Investigator's computers and on NPS computers at YNP. Spreadsheets with all eruption times, statistics, and graphs are also available.

GEOMORPHOLOGY

Project title: Holocene and Modern Geomorphic Response to Fires, Floods, and Climate Change in Yellowstone National Park: Natural and Anthropogenic Influences on Stream Systems

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Additional investigator(s): Dr. Paula M. Watt

Objective: To provide a long-term perspective on the geomorphic impacts of the 1988 Yellowstone fires, we are investigating Holocene sedimentation in northeast Yellowstone, using post-1988 fire-related events as a guide for interpreting alluvial fan stratigraphy. Comparison of the timing of fire-related events with climate proxy records elucidates the relative controls of climate, fire, and intrinsic geomorphic thresholds on alluvial systems. We are also documenting extreme floods of the last ~300 years and their effects on valley floor landscapes of northeast Yellowstone. Recent changes in stream channels seen through analysis of airphotos, historical photos, floodplain stratigraphy, and resurveying are evaluated in the context of flood history, riparian vegetation, ungulate browsing, and intrinsic characteristics of basins and channels. We are also studying a 1950 dam failure at Cooke City, MT that deposited acidic, metals-rich mine tailings along the Soda Butte Creek floodplain.

Findings: Our study of the geomorphic response to fires is largely complete, and shows that fire is both an important catalyst for landscape change and is strongly controlled by climate on 100-1000 yr. time scales. Ongoing study has identified major floods in the Lamar River system in 1918, the early 1870s, and possibly near 1800. These floods had much greater peak discharge than the 1996 and 1997 floods (the largest in gauge records), and their extensive dry gravelly deposits have lasting impacts on stream channels and valley floor ecosystems. The 1950 tailings dam break produced extreme discharges but had short duration, thus causing little erosion. However, tailings deposits along Soda Butte Creek have significant copper and lead content, impact floodplain vegetation, and continue to be eroded into the channel, adding to mining-related metal pollution.

GEOPHYSICS

Project title: Absolute Gravity and Crustal Deformation in the Yellowstone Caldera

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Additional investigator(s): Dr. David McAdoo

Objective: The primary objective of our study is to establish temporal absolute gravity and height changes in the Yellowstone caldera, and apply these measurements to constraining models for the sub-surface volcanic and magmatic sources of the caldera. Measuring the change in gravity-to-height ratio for volcano monitoring has been shown to be a powerful tool in evaluating caldera dynamics in many studies. Our gravity/GPS measurements will be integrated with regional GPS and/or SAR interferometric observations of deformation to model the sub-surface sources of the inflation/deflation of the resurgent domes (Sour Creek and Mallard Lake) within the caldera.

Findings: Our 2000 campaign was successful in obtaining eight A-10 and four FG-5 absolute gravity (and complimentary GPS) observations at our proposed stations that were suitable for measurements. We obtained a complete transect across the Sour Creek resurgent dome. This transect coincides with a long-established leveling line that is at the crux of validating inflation/deflation of the Sour Creek dome. We obtained data at two sites along the Elephant Back fault zone. This area may act as a sub-surface conduit between the Sour Creek and Mallard Lake domes. Interferometric SAR data has shown that this area experiences vertical deformation. We obtained a very intriguing FG-5 dataset at Old Faithful which shows a 10-15 microgal increase over a day which we believe is attributable to a real hydrothermal signal associated with the Old Faithful geyser. All of the data has been reduced. We are currently working on a kinematic solution for the Old Faithful GPS data, but our main efforts are in investigating the source of the gravity signal at Old Faithful.

GIS

Project title: Assessment of Land Cover Changes in the Upper Snake River Plain and Greater Yellowstone Ecosystem

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Additional investigator(s): Chris Witt

Objective: Gather vegetation ground truthing information to assess the accuracy of remote sensing image processing techniques and estimate the ecological effects of land cover change.

Findings: Accuracy and effects pending statistical analysis. Tentative results indicate the ability to confidently identify cover changes of greater than 30 percent loss or gain of cover. There is some indication of ability to identify as little as 5 percent loss or gain in cover.

Project title: Development of Algorithms to Use with Satellite Images to Assess Annual Snow Melt and Green-Up In Yellowstone National Park

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Additional investigator(s): Donay Hanson, Dr. Kathy Hansen, Dr. Richard Aspinall

Objective: This thesis is a methodology for use in conjunction with satellite imagery to determine snow cover and green up in Yellowstone National Park that can be used on a near-real-time basis. The methodology includes an Avenue Script in ArcView to perform the algorithms that accurately identify changes in snow cover and green up. They will be accompanied by a descriptive text including instructions for use and assumptions. The overall objectives of the study are to use satellite imagery and develop a method to determine percent of ground covered by snow and changes in forage quantity (green up).

Findings: The study and thesis are complete. An algorithm compatible with the Yellowstone Spatial Analysis Center, using ENVI, is being developed. Estimates of ground covered by snow (snow cover) from

two aerial reconnaissance flights were regressed against raw and calibrated pixel numbers for the five band images from the corresponding days. The linear regression model using band 4 of georeferenced and calibrated AVHRR images estimated snow cover (Adjusted R2 = 0.856, alpha = 0.001). To estimate green biomass, linear models derived by Thoma (1998) were used and ground reference data were collected during the growing season in Yellowstone National Park to evaluate the application of the model for this study area. Results of the linear model were satisfactory (Adjusted R2 = 0.592, alpha = 0.0001). The algorithm is contained in a file created to aid in the use of the models for estimation of snow cover and green biomass.

HERPETOLOGY

Project title: Status and Trends of Amphibian Populations in the Greater Yellowstone Ecosystem

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Additional investigator(s): Debra Patla

Objective: Assess amphibian status and distribution in Yellowstone and Grand Teton national parks based on: 1) surveys of all potential amphibian breeding sites in randomly selected hydrological units (7th level); 2) surveys for priority species (Northern Leopard Frog, Boreal Toad) and 3) population monitoring at sentinel sites. This project is part of the Department of Interior's National Amphibian Research and Monitoring Initiative.

Findings: Surveys were conducted in four hydrological units in Yellowstone: Arnica Creek, Buffalo Meadows, Hayden Valley, and Specimen Creek. We found breeding sites for Tiger Salamanders (11), Boreal Chorus Frog (45), and Columbia Spotted Frog (18). Priority species surveys and monitoring revealed toad breeding at 5 of 6 previously-used breeding sites. Population estimates for spotted frogs at the designated sentinel site are in progress. Survey work will continue in 2001, in partnership with NPS's Greater Yellowstone Network Inventory & Monitoring Project.

HYDROLOGY

Project title: Dartmouth College Earth Science Field Methods

Principal investigator: Dr. Xiahong Feng
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Additional investigator(s): Carl Renshaw

Objective: This is one segment of the three-month course of Field Methods in Geology and Environmental Studies taught by the faculty of Earth Sciences at Dartmouth College. The objective for the Yellowstone segment was to teach students a number of field methods used in surface hydrology and geochemistry.

Findings: The part in stream geochemistry involved sampling stream and hot spring waters for analysis of alkalinity, calcium and chloride concentrations. The sampling was designed to examine the effects of bedrock lithology and hot spring input on stream chemistry. We found, as expected, that stream water near hot springs was high in alkalinity and chloride. Stream water running over limestone bedrock was high in alkalinity and calcium. Students were able to use simple mixing models to calculate the contribution of hot spring water to various streams inside Yellowstone.

Project title: Rocky Mountain Regional Snowpack Chemistry Monitoring

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Additional investigator(s): John Sacklin, Craig McClure

Objective: The quality of atmospheric deposition from both regional and local emissions throughout Yellowstone is being monitored by the USGS and the NPS. Previous studies have indicated a correlation between snowmobile use and concentrations of ammonium and sulfate in annual snowpacks in the park, and large regional pollutant-emissions sources exist in areas near the park. Thus, the spatial extent and concentrations of the deposition to annual snowpacks of acids and other compounds associated with fossil-fuel combustion are being monitored. Results from the year 2000 study will be compared to established chemical baselines in the Greater Yellowstone Area from the period 1993-1999.

Findings: Snowpacks representing high- and low snowmobile use at three locations in Yellowstone, and snowpacks at nine additional locations representing regional deposition to other sites in and near the park, were sampled during March 2000 to measure wintertime concentrations of selected inorganic chemicals. Snowpack samples representing most of the winter precipitation were collected at about the time of maximum annual snow depth. Concentrations of inorganic compounds in snow samples during 2000 from pairs of sites located directly in and off snow-packed roadways used by snowmobiles compared consistently to concentrations in samples collected in previous years at the same pairs of sites, except at the West Entrance where unusually early snowmelt occurred and limited sampling. Concentrations of ammonium and sulfate were higher for the in-road snow compared to off-road snow for the other two pairs of sites (Old Faithful and Sylvan Lake).

Project title: Impacts of Natural and Human Disturbance on Stream Morphology, Microhabitats, and Riparian Ecology

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Additional investigator(s): Jim Rasmussen

Objective: This study examines the influence of disturbance (specifically fire and mining) on stream habitats and riparian vegetation. We have examined the distributions of heavy metals and woody debris in burned and unburned and mined and unmined streams at spatial scales of meters to watershed-wide and at temporal scales ranging from daily to annual.

Findings: Woody debris in Soda Butte and Cache Creeks appears to respond to floods and hydraulic controls in the manner of sediments. Upper portions of the basin show little change in woody debris distributions, despite large floods in 1995, 1996, and 1997. This is because much of the debris spans the channels and cannot be moved effectively, which makes the stream transport limited. In intermediate channels (3rd order), debris input and transport appear to be in equilibrium, with inputs equaling transport out of the system. As one moves downstream (4th and 5th order streams), the stream appears to become supply limited, with transport capacity exceeding the supply coming into the stream. The burned Cache Creek drainage has more woody debris, in keeping with the increased bankside supply derived from the 1988 fires. However, first order channel morphology in 40 tributaries of Cache Creek (burned) and Pebble Creek (unburned) show no significant differences, indicating that stream morphology has recovered from any post-fire changes in these headwater areas. The only streams that display significantly different morphology are channels where check dams were emplaced to reduce erosion after the fires. These check dams appear to accomplish the opposite of what they are intended to do; disturbing the natural system and preventing full recovery to pre-fire conditions.

LIMNOLOGY

Project title: The Biogeochemistry of Sublacustrine Geothermal Vents in Yellowstone Lake

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Additional investigator(s): Carmen Aguilar, James Maki, Robert Paddock, Russell Cuhel

Findings: We did not pursue any fieldwork in 2000, but will be conducting ROV operations on the Yellowstone Lake in 2001-2003. These studies will continue to focus on the microbiology and biogeochemistry of the geothermal systems in Yellowstone Lake.

MAMMALOGY

Project title: Fecundity and Fawn Mortality of Northern Yellowstone Pronghorn

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Moscow, ID 83844-3051

Objective: The purpose of this research is to obtain an estimate of annual fecundity and offspring survival of pronghorn females, and to describe the age schedule of fawn mortality. In May through June 2000, we located collared females daily, or as frequently as possible, to record pregnancy status and birth date. We captured fawns of these females to record limb length and body mass, and to attach solar-powered ear tag transmitters. We checked radio signals daily and attempted to make daily observations of females whose fawns we were unable to catch. In the final two weeks of August, we counted all pronghorn, including all surviving fawns.

Findings: 1) Pregnancy Status: Of the 26 females that we observed during the fawning season, we classified two as not pregnant. It is possible that these females lost fawns early, before we first located them. The minimum pregnancy rate is 24/26 = 92.3 percent; 2) Birth Dates: We obtained good estimates of the birthing dates for 10 females that gave birth between 22 May and June 10. The median date was May 30; 3) Birth Mass: We weighed 11 fawns. The median age at capture was three days. Mean adjusted birth mass [Mass at capture - (days age at capture x 0.2446)] was 3.12 kg; 4) Birth Sex Ratio: Of the 13 fawns that we captured, or observed closely enough shortly after birth to ascertain sex, five were male and eight were female. In the late summer count of 35 surviving fawns, 17 were male, 16 were female, and 2 were of unknown sex; 5) Fawn Survival: We fitted 12 fawns with radio transmitters, and knew the fates of two others. All 14 fawns died between 1 and 25 days of age. The median age at death was 14 days. We recovered three radio ear tags at the mouths of coyote dens; 6) Adult Mortality: Seven radio collared females died. The death rate of adult females in 2000 was 7/27 = 25.9 percent. This was significantly greater than the rate of the preceding year, which was 1/24 = 4.2 percent. Five of the seven deaths occurred with three weeks of birth and only one of these seemed attributable to other than external causes. Four females died on winter range. We saw one of these killed by a coyote and the deaths of the other three are consistent with coyote predation. It may be that there are a few coyotes on winter range that have developed the ability to kill adults and have their greatest success at this when mothers attempt to defend young fawns; 8) Location-Specific Survival of Fawns: Late August counts revealed that fawn recruitment was much higher in the eastern portion of summer range than elsewhere. These data hint at the possibility that fawn recruitment on wolf territories is higher than recruitment off wolf territories.

Project title: Carnivore Detection Survey

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Additional investigator(s): Kerry Halligan, David Bopp

Objective: 1) Assess several methods to inventory and monitor medium-sized carnivores: weasels, otter, wolverine, marten, fisher, lynx, bobcat, mountain lion, fox, coyote, and gray wolf; 2) Examine various habitat and landscape characteristics related to their presence/absence; 3) Conduct presence/absence surveys in Yellowstone National Parks and surrounding wilderness areas.

Findings: 1) During the winters of 1990 through 1997 we conducted detection surveys and evaluated three methods: hair snares, remote camera stations, and snow track transects. Their utility as estimates of presence, distribution, and abundance were evaluated, as well as their cost, maintenance, reliability, precision, and bias. Response to hair snares and camera stations were variable locally and between years. Hair snares have the exceptional advantage of providing DNA and potentially identifying individuals, but has the disadvantage of relatively high maintenance and cost and provided unreliable results from the analysis of hair characteristics. Camera stations, like hair snares, performed well in adverse weather and can identify individuals, but suffer from avoidance bias by several resident species. Camera stations were costly in terms of expense and maintenance. Snow track transects identified four species not detected by other methods and were simple, low cost, and low maintenance. They provide precise habitat information, whereas camera stations and hair snares are baited with food and scent lures which bias results concerning habitat use. Snow track transects allow researchers coverage of large areas and habitat types and can provide valuable information if scats are found and if DNA is successfully extracted. The reliability of species identification from snow track transects is a major disadvantage due to poor climatic conditions and the similarity of many species' track characteristics. Although the specifics of objectives and logistics should dictate use of these methods, we suggest a variable combination of all three methods for determining presence and distribution. All methods have significant problems, especially when inferring abundance. Determining relative habitat use from snow track transects proved reliable and matched that known from previous studies. We prepared a manuscript on the evaluation of three detection methods for medium-sized carnivores. This project resulted in the confirmation of fisher in the Yellowstone Ecosystem. Efforts in year 2000 focused on further analysis of data and preparaton of manuscripts for publication.

Project title: Study of Grizzly Bear Behavior and Genetics

Principal investigator: Dr. Steven French
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Additional investigator(s): Marilynn French

Objective: Continuation of a long term study of grizzly bear behavior and the use of nuclear DNA markers to determine genetic diversity and population estimates.

Findings: Thirty-four new nuclear DNA microsatellites have been developed from a grizzly bear genome and the genetic profiles for 155 grizzly bears have been determined.

Project title: Development of Aerial Survey Methodology for Bison Population Estimation in Yellowstone National Park

Principal investigator: Dr. Robert Garrott
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Additional investigator(s): Steven C. Hess, L. Lee Eberhardt

Objective: The overall objective of this study is the development of aerial survey methodologies for statistically rigorous estimation of the bison population in the Yellowstone area that will that will have sufficient power and precision to detect demographic trends. These methodologies will allow NPS resource managers to conduct aerial surveys that provide scientifically defensible population estimates to address ecological conditions and epidemiological management issues of Yellowstone bison.

Findings: The survey methodology we are developing quantifies a sampling universe and sampling units with computer geographic information systems, standardizes search effort, employs a stratified sampling design which accounts for undetected animals, and uses an aircraft global positioning system to record data locations. Including seasonally occupied areas outside YNP boundaries, 76 survey units with area of 2,339 km2 comprise the entirety of our designated survey extent, roughly equivalent to 26 percent of the area of YNP. The same survey units and total extent are used both in winter and in summer, but survey units have different strata designations for each season. During winter, 52 percent of the entire survey area is designated to be in the high density stratum, while in summer, 41 percent of this area is in the high density stratum. For this research, we enumerated all survey units in the seasonal high density stratum during each survey, and enumerated the low density stratum over the course of each season from February 1998 until August, 2000. Concurrent intensive ground surveys, or 'double sampling', in the Madison-Gibbon-Firehole areas and the Northern Range in winter, and Hayden Valley in summer were used to estimate the magnitude and variability in detection probability during specific aerial surveys. In comparing these simul-

taneous ground and aerial surveys primarily in winter, only 80.7 percent of the groups were detected on average from aircraft, although 93.7 percent of individual bison were detected. During the summer breeding period, as much as 70 percent of the entire bison population is aggregated in significantly larger, highly visible groups in Hayden Valley than observed during winter. Conducting surveys during this time may provide for both higher detection probability and smaller spatial extent than in winter, when bison occupy a larger area, and occupy thermal habitats that have unfavorable background color. We found detection probability to be relatively high in comparison to aerial surveys of other species, although many small groups and solitary bison were not detected from aircraft, which biases population estimates downward. Preliminary results show that low variability between counts and high detection probability provide population estimate with an overall coefficient of variation of roughly 8 percent. We are currently using Monte Carlo simulations to evaluate the efficiency of different stratified sampling designs. Our field research is now completed and we are conducting final analyses and manuscript preparation. We presented preliminary results at a national conference of The Wildlife Society in Nashville, Tennessee in September 2000.

Project title: Some Population Characteristics of the Yellowstone National Park Bison Herd. 1996–2001

Principal investigator: Dr. Peter Gogan
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Additional investigator(s): Edward M. Olexa, Kevin M. Podruzny, John A. Mack

Objective: To determine basic population parameters of Yellowstone National Park bison.

Findings: Data analysis continues and population modeling has begun.

Project title: Seasonal Habitat Selection by Bison in YNP

Principal investigator: Dr. Peter Gogan
Contact info: see above

Additional investigator(s): Edward M. Olexa, John A. Mack

Objective: To determine habitat selection by bison relative to habitat variables such as to habitat conditions such as topography, fire history, vegetative cover types, and geothermally-heated soils.

Findings: Radio-tracking ended in fall 2000. Data analysis and report preparation have been ongoing since that time.

Project title: Population Dynamics of the Yellowstone Grizzly Bear

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Additional investigator(s): Mark Haroldson

Objective: To describe the population trend from threatened status to recovery and approximate stability.

Findings: These data includes information collected by the Interagency Grizzly Bear Study Team (members include USGS-BRD, YNP, WYDGF, IDFG, MTDFWP, USFWS, USFS) for the entire Greater Yellowstone Area. Data obtained within YNP is not broken out separately): Fifty-four individual grizzly bears were captured a total of 65 times during the 2000 field season in the Greater Yellowstone Area (GYA). Thirty-two captures were new individuals that had not been previously marked. Twenty-seven captures of 24 bears were the result of management trapping efforts. Twelve of these instances resulted in relocation of the nuisance bear(s). Five management captures resulted in the removal of nuisance individual(s). A total of 1,039 aerial radio-locations were obtained from 84 individual grizzly bears radio-monitored during all, or a portion of the 2000 field season. Twenty-eight of the grizzly bears radio-monitored were adult females.

Two rounds of observation flights were conducted as part of our effort to count unduplicated females with cubs-of-the-year and document distribution of females with young (cubs, yearlings, or 2-year-olds). The first round of flights began June 5. Thirteen grizzly bears were observed in 10 groups during 47 hours of flying before we curtailed flights due to bad weather and poor bear sightability. The second round of flights began on 17 July. A total of 108 grizzly bears in 71 groups were observed during 79 hours of flying. Twenty-two females with young were observed during observation flights. Seven of these were initial observations of unduplicated females with cubs-of-the-year. Thirty-seven unduplicated females with cubs were identified during 2000. A total of 72 cubs were observed during the initial sightings of unduplicated females. Nine single cub litters, 21 litters of twins, and 7 litters of triplets were observed. Mean litter size was 1.9. Unduplicated females with cubs were observed in 14 of 18 Bear Management Units (BMU) within the grizzly bear recovery zone. Females with young were documented in all 18 BMUs. We documented 19 known, 1 probable, and 3 possible man-caused grizzly bear mortalities during 2000; 5 were management removals. Thirteen of the known and probable man-caused mortalities were related to big-game hunting activity within the GYA. Three known and 3 probable natural mortalities were documented. An

additional 4 grizzly bears (3 known and 1 possible) that died from unknown causes were discovered.

Project title: Food Habits and Habitat Use of the Yellowstone Grizzly Bear

Principal investigator: Dr. Charles Schwartz Contact info: see above

Additional investigator(s): Mark Haroldson, Shannon Podruzny, Doug Ouren

Objective: To determine habitat requirements for the Yellowstone grizzly bear and to document its return to free-ranging status.

Findings: These data include information collected by the Interagency Grizzly Bear Study Team (members include USGS-BRD, YNP, WYDGF, IDFG, MTDFWP, USFWS, USFS) for the entire Greater Yellowstone Area. Data obtained within YNP is not broken out separately: Surveys to determine an index of spring ungulate carcass availability were conducted during May. Approximately 300 km of transect routes were surveyed in four different ungulate wintering areas. A total of 44 elk, 4 bison, and 1 mule deer carcasses were observed for a rate of 0.13 ungulate carcasses/km. These results indicate a relatively small number of winter-killed ungulates were available to bears during spring 2000. Surveys for numbers of spawning cutthroat trout and their use by grizzly bears were conducted from mid May to early August on tributary streams to Yellowstone Lake. Numbers of spawning fish were average in most streams surveyed except those in the West Thumb area. Three hundred and eighty-six hair samples suitable for DNA analysis were collected near spawning streams. These samples are used to estimate of the number of individual grizzly bears that fish for spawning cutthroat trout. Results of the DNA analysis are expected by late spring 2001. Eighty-five individual grizzly bears have been identified visiting spawning streams from hair samples obtained between 1997 and 1999. Surveys of 18 whitebark pine cone productivity transects distributed throughout the GYA were completed during July. One transect was not read because of fire activity that closed the portion of the Gallatin National Forest where the transect was located. Mean cones per tree for the read transects were 5.7. This year's poor whitebark pine cone crop was offset partially by last year's near-record cone production. Grizzly bears throughout the GYA used last year's cones well into fall 2000. A total of 129 grizzly bear observations, including 10 family groups, were recorded at 17 of 46 (37 percent) of the known and/or suspected insect aggregation sites identified through 1999. Grizzly bears were observed digging in talus, presumable for moths, at two additional high elevation sites during 2000.

Project title: Black Bear Demographics in Yellowstone National Park: Their Interrelationship to Other Carnivores, Habitats, and Humans

Principal investigator: Dr. Charles Schwartz
Contact info: see above

Additional investigator(s): Mark Haroldson, Kerry Gunther, Glenn Plumb

Objective: 1) Determine patterns of habitat use, food habits, activity patterns, movements, and home

range size for a sample of randomly captured black bears; 2) 2. Evaluate the temporal and spatial patterns of habitat use of sympatric grizzly and black bears.

Findings: Three adult black bears (1 male, 2 females) were captured and fitted with Global Positioning System (GPS) collars during the fall of 2000. Seventeen aerial locations were obtained from these bears before they entered their winter dens. Detailed data on the movements of these bears will not be available until the store-on-board collars are retrieved during the summer of 2001.

Yellowstone Grizzly Bear Investigations for 1995-1999 are now available at http://nrmsc.usgs.gov/research/igbst-home.htm. The 2000 Annual Report will be available by mid summer 2001.

MICROBIOLOGY

Project title: Survey of Yellowstone Hot Springs for Green Sulfur Bacteria

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Additional investigator(s): Dr. David Ward, Dr. Ulrich Nuebel, Mary Bateson

Objective: 1) To survey selected hot springs in Yellowstone National Park for the presence of green sulfur bacteria (GSB); 2) To further characterize and possibly isolate organisms whose 16S ribosomal gene sequences indicate that they may be deeply branching relatives of green sulfur bacteria from selected Yellowstone hot springs. We sought to apply powerful molecular techniques to determine whether we could detect the presence of Chlorobium or other green sulfur bacteria in hot springs in the Mammoth region or in other Yellowstone hot springs that might provide a suitable habitat.

Findings: We designed primers specific for an 800 base pair region of the 16S ribosomal gene and used PCR amplification to screen for the presence of green sulfur bacteria in DNA extracted from 18 different locations in 11 different hot springs. Seven of these hot springs were in the Mammoth Hot Springs region. We also tested Octopus Spring and Mushroom Spring, because the Ward lab has previously detected 16S rRNA genes (E-type) that are closely related to those of green sulfur bacteria (GSB) in these sites. Finally, we tested several locations in a large unnamed pool East of Artist Paint Pots and in effluent from an unnamed hot spring in the Mud Volcano region. These were selected because they had the proper temperature and pH for previously identified thermophilic GSB even though the sulfide content was low. We detected GSB 16S rRNA sequences only in the hot spring from the region East of Artist's Paint Pots.

We subsequently enriched green sulfur bacteria from two different locations in this pool. Two enrichments grow at 36.5C, but a third grows well at 44C. Spectrophotometric analysis of the photosynthetic pigments and PCR analysis with GSB specific primers both confirm that each of these enrichments contains green sulfur bacteria. We have not yet obtained isolates from these enrichments but plan to do this. We also obtained GSB enrichments from two different locations in the Mud Volcano region, one at 36.5C, and one at 44C. Again, the identification was confirmed by both spectrophotometric analysis of the pigments and by amplification with GSB-specific primers, even though we were not successful in detecting the GSB sequences in DNA extracted directly from the hot springs without prior enrichment.

We are working to improve the sensitivity of our PCR analysis since we have still not succeeded in detecting the presence of GSB organisms in three of the four sites from which we subsequently obtained positive GSB enrichments. Obviously the organisms are present, but our assay is not sufficiently sensitive to detect them. In addition, since our enrichments were more successful than our molecular screening, we

intend to collect fresh samples from the Mammoth Hot Springs region and attempt GSB enrichments on these samples.

We have cloned an 800 bp region of the GSB 16S rRNA from each of these enrichments as well as from the DNA of the single site in which we were able to detect GSB sequences directly. Mary Bateson sequenced 40 some clones from the latter site. Comparison with previously sequenced 16S rRNAs using the BLAST tool of GenBank confirms that these are indeed green sulfur bacteria, but that their sequences are not identical to those of any of the organisms in the data base. We will be continuing our sequencing and analysis of these genes.

Project title: Bacteria Living at Low pH and High Temperature

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Objective: The main objective is to examine high temperature and low pH springs to find new (i.e. previously undescribed) hyperthermophiles and develop methods to isolate and culture them. There are three specific objectives: 1) examine and quantify existing microbes in selected springs for site reference purposes (phase contrast and epifluorescence light microscopy); 2) analyze and assess low pH high temperature habitats for the presence of microbes defined by their structural features in the electron microscope using scanning and/or transmission electron microscopy; 3) study the growth and growth rates of species in nature by observation of organisms attached to immersed slides. Physical identification of the physiological state and numbers of organisms present at a given site will allow us to design media and establish conditions used for the isolation of microbial cultures.

Findings: A previous examination of sampling sites for microbes provided the result that DNA was present in small objects regarded to be microbes. There were various shapes and arrangements of these seen by light microscopy. At Amphitheater Springs, rod-shaped organisms were present at all temperatures examined but the numbers increased at lower temperatures. Round Sulfolobus-like organisms predominated at Moose Pool (Mud Volcano area) and Great Sulfur Spring (Hayden Valley-Crater Hill area). In media prepared for isolation of organisms in aerobic conditions we found growth at lower temperatures (55 C) in Great Sulfur Spring samples. We examined several sites by electron microscopy and found a diverse collection of organisms from several sites including Frying Pan Spring, Amphitheater Spring, and Sulfur Caldron. Various sizes and arrangements of rods and spheres were the forms seen. These organisms will be isolated in prepared laboratory media.

Project title: Transition Between Lithoautotrophy and Chemoheterotrophy in *Sulfolobus* Species

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Objective: To determine the factors which regulate the metabolic status of hyperthermophilic archaea and bacteria in situ. To investigate methods for recovery of viable cells.

Findings: The effect of sample pH, sample concentration, and sample ultrafiltration was examined on the recovery of viable cells from geothermal sites at various locations in the park.

Project title: Molecular Ecology of Photosynthetic Hot Spring Bacteria that Resemble *Heliothrix Oregonensis*

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Objective: The purpose of this ongoing project is to locate, describe, and compare microbial mat communities in Yellowstone National Park that contain deep red-colored layers dominated by novel red filamentous bacteria. Previous studies, both in our lab and in the labs of collaborators, have demonstrated that red layer bacteria in one Yellowstone community are photosynthetic and use unique molecules to harvest light for energy. Taken together, these data strongly suggest that red layer bacteria are related to, but distinct from, orange filamentous bacteria *(Heliothrix oregonensis)* found in some Oregon hot springs. *Heliothrix* represents an unusual member of the Green Non-Sulfur (GNS) lineage of bacteria, a lineage believed to represent one of the most ancient groups of life on earth. The goal of our work is to use molecular methods (e.g. DNA sequence analysis) to a) better address the identity of the red layer bacteria within each separate community; (b) to compare GNS sequences from different red layer communities to better understand variation, selection, and transfer/origin of these related bacteria; and (c) to improve our understanding of diversity in this evolutionarily important lineage of bacteria.

Findings: To date, we have located approximately twelve distinct red layer communities in Yellowstone in terms of one or more of the following categories we have measured: temperature, pH, photosynthetic pigment properties, or geographical location. In the past year, we specifically surveyed the Shoshone Thermal Basin, adding one new red layer community sample to our target study. Our six-member party backpacked

in. We also surveyed the following thermal areas, all on foot: Potts (escorted, no samples removed), Sentinel Meadows (one sample removed from Sentinel Pool, beyond Mound Spring), and White Creek (no samples removed). We re-sampled four key red layer sites to track potential variation over time: Hillside Springs, Fairy Springs, Spray Geyser, and Imperial Geyser. Previous sampling at Imperial was performed at the main pool. This year, we surveyed and sampled downstream run-off as no red layer was visible in the main pool (likely because the temperature and thermal activity had noticeably increased). We described similar increases at Spray Geyser where last year's flourishing mat had also diminished appreciably.

We are currently using DNA sequencing methods to describe and compare 16S rRNA gene sequences from red layer sites. In 2000, we specifically completed DNA analyses on two key sites (Hillside and Witch Pond), observing, as hypothesized, large numbers of novel GNS 16S rRNA sequences. While our approach is designed to preferentially target red filamentous bacteria using physical separation methods and specific DNA probes, we have also isolated and described many non-red bacterial sequences, each of which is novel. All sequences have been or will be submitted to GenBank. It should be noted that much of the Hillside analysis was completed by eight undergraduates in the context of a research-driven course I teach in molecular biology.

In September, I began three years of NSF funding (Microbial Observatory/Research at Undergraduate Institute Grant) and hired a full-time technician to work on this project. In addition to continuing DNA analyses, we have constructed an extensive public website and database about this work (www.wou.edu/las/natsci_math/biology/boomer/ALLRESEARCH/rlmocover.html). Databased information includes the following: site information over time, pigment analysis, microscopy, and DNA sequence information (including GenBank Accession information, BLAST homology searches, and phylogenetic trees). This grant also supports education, specifically funding research-driven coursework and educational outreach directed at the public schools, all based on analyzing materials from this project (see website above).

Project title: Recognizing the Signature of Hyperthermophilic Biofilms: Geyserites, Epithermal Deposits and Ancient Cherts

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Objective: Identify the processes by which hyperthermophilic biosignatures are formed and preserved in the geologic record. Establish framework within which the paleobiology, paleoecology, and paleoenvironments of hyperthermophilic communities can be recognized.

Findings: Determined through a combined microscopy and phylogenetic analysis of hyperthermophilic biofilms that the distribution of distinct hyperhtermophilic communities could not necessarily be correlated with the distribution of specific morphotypes of sinter. Began testing working hypothesis that character-

istics of microbial biofilms are related to specific microenvironments within thermal springs and geysers.

Project title: Research Experience for Undergraduates: Yellowstone National Park Field Trip, Summer 2001

Principal investigator: Dr. Anne Camper
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Additional investigator(s): Darla Goeres, Melissa Cahoon, John Neuman

Objective: The Research Experience for Undergraduates program at the Center for Biofilm Engineering, Montana State University recruits talented students in various science, math and engineering disciplines to spend 10 weeks in Bozeman conducting biofilm research, learning effective technical communication skills and debating ethical issues that arise in technical fields of work and study. Yellowstone National Park serves as the perfect location to debate the ethics of harvesting microorganisms from natural environments. The students spent two days in the park observing wild type biofilms and discussing current biofilm research being conducted in the park.

Findings: The trip to Yellowstone Park increased the students' appreciation for field research. Viewing biofilm in a natural environment demonstrated the complex ecology associated with a living biofilm better than any bench-top laboratory system. The students left Yellowstone with a better understanding of the issues surrounding research in a national park.

Project title: Long-Term Effects of UV Radiation on Biodiversity and Photosynthetic Competence of Hot Spring Bacteria

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Additional investigator(s): Tracy Norris, Erich Fleming

Objective: To determine if 1-3 month UV exclusion resulted in a change in the species composition of microbial mats in hot springs and whether photosynthetic competence of the cyanobacteria was affected.

Findings: The objective of this study was to determine whether the long-term exclusion of UVR in hot spring microbial mats resulted in an alteration in microbial community composition, such as a population shift to more UV-sensitive species. Over a 1–3 month period microbial mats in three alkaline geothermal streams in Yellowstone National Park were covered with filters that excluded or transmitted UV radiation (UVR). Over some, 25 percent transmission neutral density screens were also used. For mats in the 40–45 C range there were no significant changes in species composition during the summer with or without high or low UV radiation, as assayed by DGGE after PCR amplification of 16S-rRNA genes using general Bacterial and cyanobacterial primers. Although the composition of these microbial communities appeared to be stable, surface layers of cyanobacteria protected from UV radiation (high or low) were not as competent photosynthetically as those maintained with UVR. This decrease was expressed as a loss of the ability to perform at a maximum rate under UVR + visible irradiance. However, even UV-acclimated cyanobacteria performed better when UVR was excluded. It is probable that the large differences observed reflect changes at the level of gene expression rather than changes in species composition.

Project title: Research Experience for Undergraduates: Day Trip to Yellowstone National Park

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Additional investigator(s): Dr. Warren Jones, Darla Goeres

Objective: The Research Experience for Undergraduates program at the Center for Biofilm Engineering, Montana State University recruits talented students in various science, math and engineering disciplines to spend ten weeks in Bozeman conducting biofilm research, learning effective technical communication skills and debating ethical issues that arise in technical fields of work and study. Yellowstone National Park serves as the perfect location to debate the ethics of harvesting microorganisms from natural environments. The students spent two days in the park observing wild type biofilms and discussing current biofilm research being conducted in the park.

Findings: The trip to Yellowstone Park increased the students' appreciation for field research. Viewing biofilm in a natural environment demonstrated the complex ecology associated with a living biofilm better than any bench-top laboratory system. The students left Yellowstone with a better understanding of the issues surrounding research in a national park.

Project title: Ectomycorrhizae of Thermal Soils

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Objective: To determine effects of thermal alteration of soils on ectomycorrhizae (EM) of lodgepole pine.

Findings: EM communities of pine in thermal soils are significantly different from those in non-altered soils. EM Fungi in thermal soils are not unique to these soils, but are minor players in undisturbed systems, and selected for by thermal alteration.

Project title: Isolation and Characterization of Microorganisms Extremely Resistant to DNA Damage

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Objective: Very little is known about strategies adopted by extremophiles to maintain the integrity of their genetic material in very hot environments. To survive, all cells must continuously and accurately repair lesions to their DNA caused by environmental stress. For hyperthermophiles, additional damages are inflicted on their DNA by exposure to elevated temperature. In preliminary studies, we have found that the hyperthermophile Pyrococcus furiosus is extremely resistant to ionizing radiation. Based on these observations we propose the following hypothesis: "Hyperthermophiles resistance to ionizing radiation is due to their unusual ability to repair extensive heat induced DNA double-strand breaks which occur at a much higher rate at elevated temperature. Therefore, hot environments should also be a prime source of highly radioresistant microorganisms."

The proposed project addresses the following questions: 1) What are the highest temperatures under which genome integrity can be maintained? 2) How do hyperthermophiles protect and repair their DNA? To address these questions, we will isolate and characterize novel and highly resistant thermophiles from hot springs in Yellowstone National Park containing elevated radon levels, and exposed to high fluxes of solar radiation. In addition, we will use extreme UV- and g-irradiation as selective pressure during enrichment to eliminate competing radiation sensitive microorganisms. We will investigate the accumulation of DNA lesions from exposure to sublethal doses of radiation and the kinetics of removal of those lesions with the most radiation-resistant isolates. We will assess the performance of the new isolates under simulated space conditions at NASA Goddard Space Flight Center and at the National Institute of Standard and

Technology Synchrotron facility. Long-duration tests will determine the performance limits of the isolates exhibiting the greatest survival potential. In addition, survival and recovery of microbial isolates will be measured after their exposure to full spectrum solar radiation during a Solar Extreme Ultraviolet Rocket Telescope and Spectrograph (SERTS) flight.

Findings: 1) Characterization of thermophilic microorganisms resistant to desiccation, hard vacuum and gamma irradiation: Fourteen strains of thermophiles with optimum temperature from 65 to 75°C were isolated from samples collected in Yellowstone National Park and the Kamchatka Peninsula in Russia. Strains were isolated following exposure of the samples to hard vacuum or gamma irradiation, as selective pressures. The isolates were characterized for their survival to gamma irradiation at doses up to 5,500 Gy. Their exposure to hard vacuum is in progress. We found that all the strains were highly resistant to ionizing radiation with D37 (dose for 37 percent survival) above 3000 Gy. In comparison, the D37 for Escherichia coli is lower than 100 Gy. In addition, the strains isolated with gamma rays as selective pressure showed an increase in survival compared to the strains isolated with hard vacuum. The 14 strains are being characterized at the molecular level using RFLP and 16S rRNA.

2) Exposure of microorganisms to space vacuum and EUV during a SERTS flight. In collaboration with scientists at GSFC, we determined the survival of thermophilic microorganisms to space conditions and EUV radiation (35 to 60 nm) during a SERTS flight. Cell-holders, mirrors, aluminum filters, temperature probes and electronic components were specifically designed and built for this experiment. The two microorganisms, *Deinococcus radiodurans* and the gram negative bacterium PD3D (isolated from Yellowstone National Park, optimum growth temperature 70°C) were selected for their resistance to desiccation and hard vacuum. Cultures were filtered on polycarbonate filters to obtain about 10 e8 cells per filters (single layer of cells). Two filters were mounted on each cell holder, with one of the filters kept in the dark at all times (control for non-EUV exposure). Because of the limited amount of space on the head of the SERTS telescope, only 2 cells holders were used. Aluminum filters placed in front of the samples blocked all UV radiation except for EUVs. The SERTS telescope was successfully launched on a Terrier-Black Brant rocket on July 26, 2000 from the White Sands Missile Range (New Mexico). The rocket reached an altitude of 304 km in 283.5 seconds. The shutters stay open for 441 seconds exposing the microorganisms to space vacuum and EUV radiation. The rocket payload was recovered the same day and the microorganisms were brought back to the laboratory immediately. Following their exposure to space environment, we determined the level of survival of *D. radiodurans* and PS3D cells using cell counts. For both microorganisms, desiccation has little effect on the cell survival compared to the non-desiccated control. Exposure to space vacuum (~10 e-6 Pa) however, decreased cell survival by two and four logs for PS3D and *D. radiodurans* respectively. The most interesting result of this experiment is that exposure to EUV radiation decreased the survival of both organisms by 1 log. This is the first measurement of the effect of EUV on cell survival. We are confirming these results with synchrotron studies. We will identify the DNA lesions resulting from exposure to EUV radiation and investigate the DNA repair mechanisms involved in the repair of these lesions.

Project title: The Search for Thermophilic *Protozoa* in Yellowstone National Park

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Objective: The goal of this research is to identify thermophilic *Protozoa* in Yellowstone National Park. Research on thermophilic organisms has so far focused largely on prokaryotes. In the hot springs of Yellowstone, these efforts have been successful, discovering many novel thermophilic organisms and their gene signatures of prokaryotic nature. This suggests that thermophily may not be a highly specialized adaptation of a few obscure microbes but a widespread phenomenon not limited to the *Bacteria* and *Archaea*, but perhaps extending to the eukaryotic domain as well. The possibility that there might exist thermophilic eukaryotes, specifically protozoa, is the focus of this proposal. Needless to say, Yellowstone National Park is one of the very few environments on this planet that can be expected to harbor thermophilic *Protozoa*.

Findings: The samples were collected and transported to our home lab. We extracted DNA from several samples and are working on optimization of the PCR-aided 18ssRNA gene amplification protocol to amplify the target genes.

Project title: A Survey of *Pilobolus* from Yellowstone National Park

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Objective: 1) To obtain isolates of *Pilobolus* to examine for differences in DNA sequences and cellular short chain fatty acid composition; 2) To compare isolates from various locations by contrasting morphological characters to DNA sequences and short chain fatty acids; 3) To analyze and compare nucleic acid sequences in the various isolates to compare and contrast taxa; 4) To analyze and compare cellular short chain fatty acids in the various isolates to compare and contrast taxa; 5) To study the characteristics that can be used to identify isolates.

Findings: During 2000, isolates of *Pilobolus* were collected in Yellowstone National Park during July and August. These isolates were collected from mule deer, buffalo, pronghorn, and elk from areas near Buffalo Ford, Indian Camp, Duck Lake and Mammoth Hot Springs. All isolates have been maintained in the laboratory at Indiana University East are being used as part of larger studies to distinguish among the species of *Pilobolus*. Collections of *Pilobolus* from this project are maintained at Indiana University East. It should be noted that isolates of *Pilobolus* do not survive well under cultivation. Most isolates of Pilobolus collected in earlier years have died.

Project title: Determination of In Situ Growth Rates of Filamentous Sulfur-Oxidizing Bacteria in a Hydrothermal Vent Field in Yellowstone Lake

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Objective: We will determine the specific growth rate of cells replicating within filaments of *Thiothrix*-like, sulfur-oxidizing bacteria that have attached to metal coupon surfaces positioned in a hydrogen sulfide gradient above a hydrothermal vent. Using published conversion factors, we will describe the chemosynthetic or mixotrophic biomass production of filamentous sulfur-oxidizing bacteria attached to coupons at different hydrogen sulfide concentrations and temperatures.

Findings: During August, 2000, Mary Bay and adjacent areas of Yellowstone Lake were surveyed by SCUBA for hydrothermal vents emitting hydrogen sulfide gas. Three vent fields were located at water depths ranging from 8 to 10 meters that emitted vent fluid with sulfide concentrations ranging from 0.012 to 0.469 mg/L. A vent located at the east end of Mary Bay at a water depth of 8 meters was selected for bacterial colonization and growth studies. Stainless steel coupons were suspended from tripods positioned by SCUBA in the fluid emitted from the vent. Control coupons were suspended above the lake bottom by a tripod positioned 3 meters from the vent in an area where no hydrogen sulfide was detected. At daily intervals over an 87 hour period, coupons were retrieved by SCUBA from both sites, placed in a screw cap test tube filled with surrounding water, brought to the surface, and preserved with 0.5 percent glutaraldehyde. Upon return to the Montana State University Laboratory, the bacteria that had attached to each coupon were stained with a fluorochrome for microscopic visualization. The number of individual cells in the largest filament attached to each coupon was determined.

The motivation for evaluating the number of cells in the largest filament on each coupon is based on our assumption that this filament arose from a single cell that had been one of the early colonizers of the coupon upon exposure to the hydrogen sulfide-containing vent environment. The hydrogen sulfide is thought to serve as the primary energy source for replication of the bacteria attached to the coupon. The attached bacteria replicate inside a filament that extends perpendicular to the coupon surface. The filament elongates as the cells within the filament replicate. By determining how many cells exist within the longest filament attached to coupons recovered at daily intervals, in situ growth rates or generation times can be estimated.

To date we have identified the largest filament on one coupon retrieved after 14 hours exposure to the vent fluid and one coupon exposed for the same period of time to ambient water outside the vent field. A 203 um-long filament containing 66 bacterial cells was the largest filament on the coupon exposed to the vent fluid at a temperature of 52-54C. This equates to a specific in situ growth rate of 0.43 generations/h or a generation time of 2.3 hours over the first 14 hours of exposure. An 82 um-long filament containing 36 cells was the largest filament attached to the coupon exposed to ambient lake water at a temperature of 16C. This latter result indicates that either hydrogen sulfide is diffusing from the lake bed sediment at concentrations below the detection limits of our assay, but at concentrations sufficient to support replication of the observe filamentous bacteria in areas where vents are not apparent, or that these filamentous bacteria can replicate on an available energy source other than hydrogen sulfide.

Between March 1 and July 1, 2001, we will complete the analysis of bacterial growth rates on coupons collected at later times during the 87 hour study. We would like to deploy new coupons in the same area and conduct similar studies during the summer of 2001 in order to evaluate the reproducibility of our Year 1 data. We also want to determine whether the vent is still actively emitting hydrogen sulfide at similar concentrations as those measured during August of 2000.

Project title: Protein Comparison of Thermophiles and Oral Bacteria

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Objective: Oral bacterial microflora are extremely diverse (more than 300 different species in the normal oral cavity) and have to survive relatively large temperature and nutritional variations. Thermophilic microorganisms have been fairly well described, but no comparison has been reported with oral bacteria. It is proposed here to compare protein antigens between thermophilic and oral bacteria by immunological and electrophoretic (protein size) techniques. SDS-PAGE electrophoresis will be used to compare the sizes of proteins between representative thermophiles and laboratory strains of oral streptococci (primarily *Streptococcus mutans,* the causative agent of human dental caries). Immunological assays such as ELISA and western blots will be used to compare reactivity between antibodies to protein antigens on *S. mutans* and the thermophiles. It is anticipated that similar proteins will be observed between thermophiles and oral bacteria implying a possible common ancestry.

Findings: Bacterial colonies were isolated on both selective and non-selective petri plates. Selected colonies were propagated and stored frozen until assayed. Samples are currently being collected from human volunteers to compare to park samples. It is anticipated that more park samples will be required due to poor growth of several bacterial colony types upon secondary propagation.

Project title: Analysis of a Eukaryotic Microbial Mat Community Across Environmental Gradients in a Thermal, Acidic Stream

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Objective: Two Nymph Creek sites, defined in terms of differing light, pH and temperature, will be extensively monitored over diurnal and seasonal time periods. Contemporary analyses, including modern microscopic methods and rRNA sequencing will be used to document microbial diversity and analyze the mat's microstructure at an upstream sunny site and a downstream shaded site. Changes in macro- and microscale environmental conditions in the bulk water and through the vertical aspect of the mats will be recorded. Nucleic acid sequence-based techniques will be used to monitor population changes in situ under environmental conditions that vary in space and time.

Findings: Environmental monitors are in place to record temperature at the upstream and downstream sites (HOBO dataloggers) and to monitor PAR (photosynthetically active radiation). DNA clone libraries have been prepared seasonally and are being analyzed. Sequencing is underway on Cyanidium caldarium and other eukaryotic organisms identified from clone libraries and pure cultures.

Project title: Physiology and Geochemical Tracing of Fes/H2S Microorganisms in Subsurface Hydrothermal Systems

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Objective: To characterize the diversity of microorganisms associated with sulfide-containing or acidic hot springs. The initial phases of the study concentrated on evidence for pyrite-forming anaerobic bacteria. Subsequent work has concentrated on recovery, preservation, and molecular analysis of thermoacidophilic archaea.

Findings: In early June 2000, we assessed and sampled a total of 74 acidic outflows in three different areas of the park. These samples consisted of water plus sediment from pools having a temperature at or above 65º C and a pH value below about 6.0. This year we emphasized sampling in different regions of the park in order to look for differences in microbial populations. In addition, however, we also wanted to test possible changes in populations over time. As a result, we targeted two new areas this year (Crater Hills/Sulphur Mountain and Geyser Creek) and returned to one area sampled last year (Ragged Hills).

Sampling consisted of scooping sediment and spring fluid into sterile 20-mL glass vials, which were sealed and transported back to Cincinnati under ambient conditions. Two modifications of this technique were evaluated for their ability to increase yield of viable cells: exclusion of air from the vials, and buffering the pH at about 4. Neither technique produced dramatically different results from the simpler method of unbuffered, aerobic storage. This year we also took a few samples of hot, acid soil.

We recovered thermoacidophilic archaea from each of the three areas sampled. Overall, about 60 percent of the samples yielded colonies by direct plating. This resulted in more colonies than could be archived; as a result, we used various microbiological criteria to identify a smaller number of isolates representing the widest range of diversity possible. After applying these criteria, approximately 915 isolates were streaked for isolation, grown in liquid culture, and stored at -70 C for subsequent analysis.

The genetic diversity of isolates is being evaluated in comparison with isolates cultured in 1999 and with isolates recovered from other parts of the world in 2000. The analyses are proceeding rather slowly due to the large number of isolates on hand. However, qualitative molecular typing based on restriction of genomic DNA suggests low species diversity in the culturable populations at all park locations examined. Furthermore, this, or a closely related species or sub-species, dominates the culturable populations at other geothermal sites evaluated, including those outside North America. Quantitative relationships will eventually be assessed using DNA sequence comparisons of multiple genes, in a collaboration with Rachel Whitaker and her Ph.D. advisor John Taylor in the Department of Plant and Microbial Biology, UC-Berkeley.

Project title: Molecular and Functional Ecology of Hot Spring Photosynthetic Mats

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Objective: The project combines modern microbiological, molecular biological, and ecophysiological methods for studying the microbial ecology of photosynthetic hot spring mats. The goal is to gain a better understanding of the structure and function of the photosynthetic community in hot spring microbial mats.

Findings: Main emphasis in 2000 has been on data analysis and laboratory experiments on mat samples. A

field campaign in collaboration with Drs. Ward and Ferris during late summer focussed on obtaining further microsensor data in Mushroom Spring and Nymph Creek over a diel cycle. Studies of photosynthetic performance of various cyanobacterial isolates were conducted in the lab of Dr. Ward. Together with Dr. Nübel (Ward lab) a spectral characterization of various *Chloroflexus* type isolates as well as the spectral light distribution in mat samples was obtained. In 2001 further field measurements are planned with emphasis on dark metabolism in the mats.

Project title: Microbial Biotransformations and Ecology

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Objective: To isolate microorganisms with unique metabolic activities allowing for the transformation of C-1 compounds, polycyclic aromatic compounds, and related products from the petroleum industry.

Findings: Samples obtained from thermal features in Yellowstone National Park during August 2000 have been used as innocula for enrichment cultures. Methane and methanol were provided as sole carbon and energy sources. Cultures were maintained aerobically under mesophilic and thermophilic conditions. Isolation and characterization of microorganisms capable of growing on C-1 compounds under these conditions will continue through 2001. Microbial community structure in thermal features will be studied using soil samples that remain in a freezer repository. New enrichment cultures may be started if fresh inocula are obtained in 2001.

Project title: Bacterial Diversity of Thermophilic Photosynthetic Bacteria

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Objective: The main objective of this research is to discover and isolate laboratory cultures of anoxygenic (non oxygen-evolving) photosynthetic bacteria from thermal environments. Photosynthetic bacteria are model organisms for the study of basic problems in photosynthesis and thermophilic phototrophs are very desirable because of their thermostable photosynthetic machinery. The long-term goal of the research is to

probe photosynthetic diversity in hot springs of various chemistries and temperatures to determine the physiochemical limits to photosynthesis. This includes isolating and characterizing new species of photosynthetic bacteria and studying their basic biological properties including physiology, biochemistry, and phylogenetic position, in laboratory cultures. All cultures of thermophilic phototrophs from Yellowstone as well as New Zealand thermal springs have been deposited in the American Type Culture Collection (ATCC) for public access by any qualified individual. This is basic research; no commercial funding or research ties exist between this project and any for-profit organization.

Findings: Sampling was done in September 2000. However, our timing was not too good this year as two of the three days available for sampling were spent at the Old Faithful Inn because of a heavy early season snowstorm. Some sampling was done in the Mammoth Upper Terraces and in a small warm acidic hot spring along the Gibbon River near Beryl Geyser. Samples were subjected to DNA extraction and PCR amplification using a series of primers specific for proteins unique to the photosynthetic reaction center of purple anoxygenic phototrophic bacteria. This work is in progress but early results seem to confirm suspicions from enrichment culture experiments that several purple bacteria inhabit these springs. Currently, two pure cultures of purple bacteria, one from each of these two springs, has been isolated and characterized but the molecular results suggest that the diversity of purple bacteria in these springs is much greater.

Project title: Characterization of the Microbial Rhizosphere Population of Acid and Thermotolerant Grasses Associated with Hot Springs and Microbial Diversity in Thermal Soils in YNP

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Additional investigator(s): B. Inskeep, M. Burr, L. Botero, J. Christiansen

Objective: To study the diversity and identification of the thermophilic and acidophilic organisms associated with thermophilic plants located in YNP. We are also very interested in examining the diversity of the microbial community that thrives in select thermal soil locations.

Findings: We have obtained molecular evidence that some thermal soils (Temp. = 65°C to 85°C) apparently have diverse and complex prokaryotic communities. This study is continuing as we are developing new culturing techniques to cultivate maximal numbers of different prokaryotes from these soils. Physiological and biochemical characterization of these different isolates will then follow.

Project title: An Analysis of Soil Microbial Community Structure in an Evolving Thermal Soil Environment

Principal investigator: Dr. Timothy McDermott
Contact info: see above

Additional investigator(s): Tracy Norris, Lina Botero, Jon Wraith, Mohamad Etayebi

Objective: The objective of this work is to use molecular methods to analyze soil microbial community succession in response to changes in soil temperature. Investigations of the biology of hydrothermal systems have added greatly to our understanding of microbial species diversity and their evolutionary relationships. However, previous studies have generally been limited to thermal systems that are well established on the time scale of human observation. The death of lodgepole pines in this study site are indicative of a very recent expansion of the underlying geothermal plumbing. In some places temperatures as high as 80 ºC were recorded, which only six months previously were closer to 25 ºC. This study site provides us with a unique opportunity to observe changes in microbial community structure as they are occur. This work will allow us to address questions concerning the forces affecting microbial community structure, diversity and the colonization of geothermal features by thermophilic microorganisms.

Findings: A research plot was designated and thermocouple probes were inserted in the ground at specific locations within the plot to measure soil temperature at regular intervals. Temperature data collection was initiated in November 1999. Results indicate that the research plot includes an area of expanding geothermal activity. Soil samples were collected at several sites within the research area. Extraction of nucleic acids (DNA and RNA) from these samples by conventional protocols was analyzed via PCR and denaturant gradient gel electrophoresis. Clone libraries were also constructed and are being sequenced to compare the bacterial diversity of the sampling sites. Results suggest that temperature has been a selective force in thermally impacted soils as evidenced by apparent reduced species diversity from these sites. Total viable thermophiles (isolated on a variety of medi) were significantly higher in the thermally-impacted soils relative to unimpacted soils.

Project title: Phylogenetic Analysis of High-Temperature Ecosystems

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Objective: Ongoing research in the park continues to focus on the survey of microorganisms in microbial ecosystems with varying solution chemistries in Yellowstone. A molecular approach based on cloning and sequence analysis of the small sub-unit (SSU, 16S RNA (rRNA)) ribosomal gene is used to determine the

microbial composition of these ecosystems. Ongoing studies include analyses of sub-aqueous and sub-aerial systems for bacterial, archaeal, and eucaryal life.

Findings: Survey of eucaryotes in anaerobic, low temperature, and high temperature environments: the anaerobic eucaryotes so far encountered display broad diversity, in the range of new genera to kingdoms. Molecular environmental studies of the microbial constituents of the eucaryal domain of life, which includes plants and animals, has never been done at Yellowstone. We have discovered novel bacterial phylogenetic divisions through molecular biological analysis from samples obtained in the park.

Project title: Exosporial Membrane Characteristics of Thermophilic *Clostridia*

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Objective: To: 1) examine indigenous spore forming bacterial species from out-flow channels of neutral pH hot springs preferably containing calcium and magnesium (travertine type springs) ranging in temperature from 35 to 60 degrees centigrade; 2) examine culturable endospores from these species in terms of their growth characteristics; exosporial membrane morphology at dormancy, activation, throughout germination and at bacterial vegetative cell outgrowth; whether these endospores are capable of attachment to substrates or each other via cellular processes or exopolymer production; histochemically and biochemically, what materials enable these endospores to attach to substrates, other bacteria and form mats or floating islands in the absence of mature vegetative cells; the speed and stages of attachment for the spores capable of growing under laboratory culturing conditions; and the visco-elastic properties of the attachment using Atomic Force Microscopy; 3) document by electron microscopy (TEM and field emission SEM) the ultrastructure of the hot spring endospores from dormancy through germination and vegetative cell outgrowth; and 4) compare the attachment mechanisms of the endospores in the hot springs with the attachment mechanisms we have been studying in pathological *Bacillus* and *Clostridial* isolates.

Findings: We have isolated and have inculture 11-14 bacterial types isolated from Terrace Spring outflow channel; and 3 or 4 new bacterial varieties isolated from the Firehole cascades. Although each organism isolated thus far has individual characteristics (i.e. colony type, anaerobic vs. aerobic habitat, odor, color, staining characteristics, spore morphology etc.), we recently analyzed each isolate using the Vitek automated identification system, and predominantly found 4-5 isolates to be Bacillus licheniformis. In Firehole cascades, there seem to be multiple strains of Bacillus pumilus, as well as B. licheniformis. Initial studies seem to suggest that a number of strains of the same organisms seem to exist in these hot springs, but the spores from each strain have very different attachment capabilities, morphologies and exosporial membrane ultrastructure. Some endospores actually have intra-exosporial vesicles that are released at outgrowth, thereby anchoring the small immature vegetative cells to neighboring debris,and preventing them from

being swept away.

The Firehole cascades produced very different endospores, many of which could not be stained with traditional spore stains. When these unstaining spores were treated with carbol fuchsin, the spores stained deep red to bright pink, suggesting the presence of surface lipid. Ultrastructural studies showed that these spores had a lipid-like outer layer during dormancy, which peeled away during germination to reveal an underlying reactive exosporium with numerous attachment projections. Therefore, we have found that the unstainable (clear) endospores that we often observed in the higher temperature hot springs seem to have a covering lipid layer but can be stained with carbol fuchsin. Later during germination, the lipid layer either diminishes or is sequentially removed, which allows the spore to progressively stain more intensely with traditional stains.

We were very surprised to find that when all of the mechanisms of endospore attachment of these hot spring species were compared to the attachment mechanisms of the human pathogens we study, the hot spring endospores had numerous unique strategies for attaching themselves, as well as ways to make a supportive network to keep spores and newly released bacterial cells attached to one another or to a substrate. All of the hot spring endospores had the exosporial attachment structures seen in our pathogenic organisms (*C.sporogenes*, *C.difficile*, *C.clostridioforme* etc.). However, the hot spring organisms had additional spore anchoring systems that seem to be unique to these environments. In some cases, the hot spring spores made a "spider web" type network interconnecting individual spores. But in other cases the material holding the endospores together was very mucoid, and in one isolate a very heavy, ruthenium red-osmium exopolymer securely tethered the spores to other spores or a substrate. In colony cultures many of the mature vegetative cells provided the mucoid, and filamentous network to keep spores attached to the colony. However the spores alone (in the absence of living vegetative cells) could make adequate attachment structures to facilitate adherence and insure survival.

Project title: Ecology of Phototrophs in Extreme Environments: Thermal and High Iron

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Objective: We are trying to determine the distribution and role of both oxygenic and anoxygenic phototrophs in extreme environments. We are also trying to determine how these bacteria interact with each other, with other microbes, and with the physical/chemical environment. Ultimately we hope to better understand the early evolution of phototrophs and their impact on the Precambrian Earth.

Findings: This year we studied the highest temperature phototrophic microbial mats in high iron springs at Chocolate Pots. These particular mats are found around 50 degrees C. They are composed of the oxygenic *Synechococcus* and the anoxygenic *Chloroflexus*. Both of these phototrophs appear to play an important role

in this high iron hot spring. Microelectrodes were used to determine the oxygen and pH levels within the mats under light and dark conditions. The *Synechococcus* clearly produced oxygen in the light but the levels of oxygen produced in these mats were much lower than in other lower temperature mats in these same springs and in other *Synechococcus* mats studied in low iron springs elsewhere. The pH was also lower than observed in other phototrophic mats. We have found some evidence for an iron stimulation of photosynthesis in the anoxygenic phototroph, *Chloroflexus*, cultured from these mats.

Project title: Spectral Analysis of Hyperthermophile Organisms, Yellowstone National Park

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Objective: The objectives of the project are to determine whether hyperthermophiles have characteristic reflectance spectra which can be used to uniquely identify species. If such unique spectral characterization can be established, the next question is whether it persists after death and possible incorporation into sinter materials and for how long it might persist. To achieve this objective we acquire visible-near infrared spectral reflectance data at wavelength of 350 to 2500 nm of various hyperthermophile organisms growing under various temperatures as well as associated siliceous and carbonate sinter deposits. Targets include various living organisms, organisms that have died but remain in situ, and organic remains that have become entombed into sinter as well as pristine siliceous and carbonate sinter.

Findings: To date, reflectance data have been collected at Lemonade Creek, Nymph Creek, Octopus Spring, Mushroom Spring and in the Upper Geyser Basin. From analysis completed, it can be concluded that the hyperthermophilic organisms do have characteristic spectra. Reflectance data for Thermocrinis ruber, *Synechococcus* lividus, *Chloroflexus* aurantiacus , Mastigocladus laminosus and Cyanidium caldarium from Octopus and Mushroom Springs and Nymph Creek show that each has a characteristic spectra significantly different from each other. Reflectance data for siliceous sinter from Mushroom and Octopus Springs exhibits absorptions which can be attributed to organic materials incorporated into the sinter. Comparison with sinter from the Upper Geyser Basin, which lacks macroscopic hyperthermophile populations, shows that ordinary sinter does not exhibit such organic signatures.

Project title: Isolation and Characterization of Thermophilic Microorganisms

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Objective: Isolation and characterization of thermophilic microorganisms, and the purification and characterization of their enzymes.

Findings: Microbial fieldwork included two brief visits to Yellowstone in 2000 to sample a lower- temperature hot spring where we had previously obtained high levels of thermophilic *Naegleria* isolates and where there has been an increasing level of recreational hot potting (e.g., Huckleberry Hot Spring and the springs along Polecat Creek near Flagg Ranch).

We also participated at the Seventh Interagency Science Conference to present the results of our earlier *Naegleria* survey to encourage other Yellowstone researchers to consider this area of study of eukaryotic microorganisms in the Yellowstone Ecosystem. We obtained some further samples of previously studied sites (White Creek, etc.) in an ongoing study of the types and populations of the thermophilic microorganisms present in these sites. Results include: 1) The previous survey for thermophilic *Naegleria* in the Greater Yellowstone Ecosystem (presented at the 1995 Yellowstone Conference) has been published and other investigators are reporting *Naegleria*-like 16S rRNA sequences in their Yellowstone environmental samples. One can anticipate that this will be a productive new area of investigation in Yellowstone. 2) Samples from Mushroom hot spring have been shown to have significant levels of *Thermus (Meiothermus) ruber.* Previous sampling had shown that these gram negative, red pigmented bacteria, which have an optimum growth at 60 C, are often present in many of the lower temperature hot springs in the Yellowstone ecosystem (e.g., the springs upstream in the White Creek area and at Huckleberry/Polecat Hot Springs) and the lower portions of runoff channels of higher temperature hot springs. 3) Yellow, spore forming bacterial isolates obtained from Octopus, Twin Butte Overlook, and Calcite Hot Springs and postulated to be *Bacillus flavothermus* have been confirmed to be *Bacillus flavothermus* as determined from the nucleotide sequence of their16S rRNA. *Bacillus flavothermus* differs from *Bacillus stearothermophilus* and higher temperature Yellowstone isolates (e.g., *Bacillus caldolyticus*) by its ability to grow both at 55-60 and at 25-37 C. The rather broad range of growth temperature shown by *Bacillus flavothermus* may play some role in its rather widespread distribution. These isolates are also consistent with the 16S rRNA sequences and isolate obtained from the runoff channel from Octopus Spring by Dr. David Ward and his research group at Montana State University. 4) Another apparently new isolate is a member of the *Exiguobacterium* genus that has been obtained from lower temperature hot springs (30-35 C) in Yellowstone and western Colorado. These are gram positive, low G+C, non spore-forming bacteria and determination of their 16S rRNA sequence indicates that they are distinct from bacteria previously found in thermal areas.

Project title: Geochemical Constraints on the Ecology of the Deep Lineages within the Bacteria and Archaea

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Objective: To 1) determine the microbial diversity and geochemistry associated with high temperature thermal springs in YNP and 2) study the ecology of microbial communities inhabiting YNP thermal springs.

Findings: Our research in 2000 was focused on Calcite Springs and Obsidian Pool. We collected extensive geochemical and molecular biological samples along chemical and physical gradients in the springs. Additionally, enrichment culture techniques were used to isolate novel thermophilic microorganisms. Initial results indicate that the geochemistry and community structure of the springs is dynamic on a spatial and temporal scale. Our research in 2001 will focus on linking geochemical and community differences and using our cultures to understand the physiological diversity of Calcite Springs and Obsidian Pool.

Project title: Analysis of Metal Resistance in Yellowstone Bacteria

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Additional investigator(s): Dr. Barrie Johnson

Objective: Identify and characterize heavy metal resistant bacteria from thermal features within YNP.

Findings: This year's sampling focused again on the Norris area. Samples were obtained north of the main Norris geyser basin (Frying Pan Hot Spring), and south of the main Norris area near Tantalus Spring. Enrichments were performed to recover a variety of acidophilic bacteria found in these thermal features. In the laboratory, we have also supported Montana State University's Thermal Biology Institute in their analysis of thermophilic viruses, through DNA sequencing of clones they have generated.

Project title: Characterizing DNA Methylase and Restriction Enzyme Genes in Environmental DNA

Principal investigator: Dr. Richard Roberts
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Additional investigator(s): David Ward

Objective: To discover novel restriction enzyme and methylase genes in environmental DNA samples.

Findings: We collected samples in three areas:

1. White Creek (Mushroom Spring - 22 samples; Octopus Spring - 29 samples; Pine Spring - 17 samples; and several other unnamed springs along White Creek for which we made up our own names such as "Bath Tub" or "Black Squid" - 23 samples from these sources). Total samples: 91, total consumed to date: 40, (the rest are stored at -70). We collected samples of biomass (filaments or mat) from 0.5 ml to 15 ml, with most samples being 0.5, 1.5 or 3.6 ml, and also samples of sediment (mostly 15 ml or 50 ml, since these will have lower titer of organisms). Some samples are duplicates of the same material to be used for different DNA extraction techniques (i.e., Mo Bio samples are placed directly into the manufacturer's tube containing buffer and beads for bead beating). Thus, the total number of samples is higher than the number of unique environments sampled. Samples consumed: All the "Mo Bio" samples have been consumed to make DNA (using the bead beating method). This includes 13 of the samples from Mushroom Spring, 11 samples from Octopus Spring, 7 samples from Pine Spring, and 3 unnamed samples. In addition, 3 other Mushroom samples and 3 other Octopus samples have been converted to DNA.

2. Nymph Creek. Sites at several sources along the creek and three small pools (no formal names for these). 31 samples collected (some are duplicates for different DNA extraction techniques (ie, Mo Bio samples are placed directly into the manufacturer's tube containing buffer and beads for bead beating). We collected samples of biomass (filaments or mat) from 0.5 ml to 15 ml, with most samples being 0.5, 1.5 or 3.6 ml, and also samples of sediment (mostly 15 ml or 50 ml, since these will have lower titer of organisms). Consumed: 11 samples have been consumed to make DNA (the Mo Bio ones).

3. Mammoth Springs Upper Terrace area. 52 total samples collected: 22 from Tangerine Spring and 30 others from various sources. 25 samples have been consumed to make DNA (the ones labeled "Mo Bio").

Project title: Genetic Analysis of Brucella from Bison and the Generation of a PCR-Based Diagnostic System for Epidemiological and Ecological Studies

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Additional investigator(s): Dr. Regina Redman, Dr. Frank Roberto

Objective: The objectives of this work are to 1) determine the genetic complexity of *Brucella* isolates from a variety of animal hosts; 2) develop a high sensitivity PCR based diagnostic system to identify the presence of *Brucella* isolates; 3) develop a PCR based diagnostic system to track specific genotypes of the

Brucella isolates and 4) develop a PCR based diagnostic system to discriminate live *Brucella* cells from dead cells. In addition, studies will be performed to convert the diagnostic systems to field adaptable systems capable of simple and rapid data generation.

Findings: We have completed the genetic analysis of *Brucella* isolates from several animal hosts including bison, cattle, and elk. These data are currently being incorporated into a scientific manuscript. In addition, several PCR primer sets have been prepared that amplify products specifically from *Brucella abortus* isolates. Protocols have been developed for extracting *Brucella* cells from blood samples and detection using the PCR diagnostic system. This year, genotype-specific PCR primer sets will be generated for tracking isolates in the field and studies will begin to establish a diagnostic system to differentiate live and dead cells.

Project title: Microbial Physiology and Ecology: DNA Damage and Photosynthesis

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Objective: The objective of this project is to study diurnal patterns of organismal physiology (e.g., photosynthesis, DNA synthesis) in order to better understand evolution on early earth and the way organisms function in their environment today. In 1999, the focus was on the effect of two naturally-occurring DNA damaging agents on DNA synthesis rates, ultraviolet radiation and hydrogen peroxide.

Findings: In 2000, research focused on the effects of UV radiation and hydrogen peroxide on microbial mat communities in Norris Geyser Basin and Octopus Spring. We found that UV radiation enhances DNA synthesis rates during the day, which we interpret as being indicative of excision repair. However, previous work suggests that the damage may be due to UVA effects mediated through oxidative damage rather than the direct effect of UVB. Experiments adding hydrogen peroxide to sample showed an increase in DNA synthesis in response to small amounts of additional hydrogen peroxide, and a decrease in response to high levels, with another increase at even higher levels, about 1 mM for Octopus and *Zygogonium* mat. At the very highest concentrations of H2O2, DNA synthesis, of course, drops to zero, probably an indication of cell death. For all the mats studied, DNA synthesis stopped by 1 M H2O2. Prechallenging *Zygogonium* with H2O2 prior to measuring the effect of H2O2 on DNA synthesis decreased the subsequent rate of DNA synthesis. This is suggestive of an induction of catalase. Techniques for studying levels of catalase and superoxide dismutase were begun in collaboration with Vanessa Lancaster and Bob Blankenship, Arizona State University. These studies will be repeated and extended in 2001.

The effect of several drugs were tested on the effect of H2O2 on DNA synthesis. Caffeine (1 mM) increased DNA synthesis in the presence and absence of additional H2O2 in *Cyanidium, Zygogonium*. *Zygogonium* mats that were placed under UV opaque screens from September to June 26, 2000 showed a down regulation in DNA synthesis when finally exposed to solar radiation in contrast to mat that was left under the UV opaque screen.

Project title: Isolation, Identification, and Characterization of Microorganisms Living in Extreme Environments

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Objective: 1) To train and inspire undergraduate students in the field of microbiology. 2) To identify and characterize the many unidentified and novel microorganisms associated with the thermal features in Yellowstone National Park.

Findings: With monies received from the Ledford Foundation through the Appalachian College Association, I was able to bring 9 students with me to the park this past summer to learn how to do field sampling of the thermal springs. The students were able to observe and learn something about sampling techniques and laboratory techniques in molecular biology. All of the samples I obtained from the park were consumed and I will return again in the summer of 2001 to obtain more samples.

Project title: Diversity and Habitat Range of Sulfate-Reducing Microorganisms

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Additional investigator(s): Sue Fishbain, Jesse Dillon, Heidi Gough, Amy Dahl

Objective: Our research at Yellowstone National Park has focused on better defining the diversity of sulfate-reducing bacteria along environmental gradients of pH and temperature. Organisms having the capacity to respire sulfate drive a key step in the global cycling of sulfur and are likely an important biological presence in many of the sulfur-rich geothermal areas within Yellowstone National Park. A long-term objective is to better define the environmental limits of dissimilatory sulfate reduction.

Findings: The recovery of deeply-diverging phylogenetic lineages, as defined by DSR gene sequence divergence, suggests that our current understanding of this important functional group of microorganisms is incomplete. Our combined analyses of different regions throughout the park suggest that sulfate respiration is a significant biogeochemical process in many of Yellowstone's geothermal features. Study sites include Bath Lake Vista, New Pit Spring, and Rolands Well Spring within the Mammoth Hot Springs

Region, Octopus and Mushroom Springs, 2 sites at the Nymph Creek area denoted as Nymph Creek and Black Sediment Pool, 4 sites in the Washburn region denoted as Site A, B, Acid Inkpot and Inkpot, 5 sites at Norris 100 Springs Plain denoted as Sites C, D, E, Cinder and Black Spring, and Obsidian Pool and Moose Pool in the Mud Volcano area. These sites provide a wide range of temperature and pH gradients (38 degrees-91 degrees C; pH 2-8). Studies in 1999 revealed significant rates of sulfate reduction at Site C, Obsidian Pool, Nymph Creek, and Black Sediment Pool. In 2000 we refined our activity assessments at these site by amending site material with alternative electron donors (CO, lactate, acetate, hydrogen, methane), testing for substrate-specific stimulation of sulfate reduction. We observed little or no stimulation at most sites, with the exception of some stimulation by hydrogen. In order to further constrain the contribution of sulfate respiration to total microbial production we determined the total phospholipid content of sediments collected from the different study sites.

Project title: Isolation of New Hyperthermophiles and Investigations of Hyperthermophilic Biotopes

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Additional investigator(s): Robert Huber, Wolfgang Eder, Christian Rudolph, Manuela Baumgartner

Objective: Isolation of new hyperthermophiles and investigations of hyperthermophilic biotopes.

Findings: During a field trip in Yellowstone, samples from different hot springs (e.g. Nymph Lake area, Ojo Caliente, Norris Geyser Basin) were taken for microbiological investigations.

Project title: Integrated Biogeochemical Database

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Additional investigator(s): Ronald C. Rope, Peter Pryfogle

Objective: The objective of the project is to develop an Internet-accessible database that integrates microbial diversity data with geochemical information and geographical location.

Findings: We report the development of a prototype Internet accessible database and Geographic Information System (GIS) application (http://remus.inel.gov/ynphome).

Project title: Development of Harsh Environment Biosensors

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Additional investigator(s): William Apel, Diane Key, Kastli Schaller

Objective: The objective of this study is to culture thermophilic microorganisms from Yellowstone hot springs. These organisms will then be tested for the presence of various enzymatic activities and the enzymes will be isolated and purified from organisms showing high levels of activity. The enzymes will be characterized to determine how high temperatures affect their activity and stability compared to low temperatures versions of these enzymes. After the enzymes are characterized, they will be utilized for development of biological sensors.

Findings: On March 29, 2000, water and water/sediment samples were taken from the Norris Geyser Basin area. These samples were used as inoculum in defined media for bacterial enrichments. The sample identification is listed below, along with the location, temperature, and pH of each sample: BSno.1: Beryl Spring, 85.1, 7.08; BSno.2, Beryl Spring, 79.2, 7.20; BSno.3, Beryl Spring, 59.9, 2.21; CSno.1, Cistern Spring, 82.8, 5.64; CSno.2, Cistern Spring; VGno.1, Veteran Geyser, 58.5, 7.52; NNno.1, Pool below Veteran, 69.0, 4.16; NNno.2, Pool after Pearl Geyser, 82.1, 3.37; YFSno.1, Yellow Funnel Spring, 87.2, 3.87; GDSno.1, Green Dragon Spring; CRSno.1, Crater Spring, 74.1, 4.39; EGno.1, Echinus Geyser, 62.4, 6.73; SDno.1, Steamboat, 76.2, 6.75. The samples were stored in an 80°C portable Igloo heater for transportation to the lab. Temperature and pH were not taken for CSno.2 and GDS no.1 due to the sample location.

Upon returning to the laboratory, the samples were immediately placed into defined liquid or semi-solid media and incubated at either 70°C or 60°C on tabletop shakers. Three media were chosen: 1) Sulfur medium (Handbook of Microbiological Media), pH 2.5; 2) Sulfur medium, pH 4.8, or; 3) Thermus medium (ATCC no.697), pH 7.5. The Sulfur medium was modified to contain 0, 10, 100, or 1000 ppm arsenate and the Thermus medium was modified to contain either 0 or 10 ppm arsenate. After 4 days of incubation, growth was visually observed in thermus media containing 10 ppm arsenate inoculated with the following samples: VGno.1, EGno.1, BSno.2, SDno.1, NNno.2, CSno.2, and BSno.1. After 12 days of incubation, growth was observed in NNno.2 which was inoculated into sulfur media pH 2.5 1000 ppm arsenate. Growth was also observed in SDno.1, VGno.1, BSno.2, and EGno.1 inoculated into sulfur media pH 4.8 at 10 ppm arsenate. Growth was observed in YFSno.1 in 1000 ppm arsenate sulfur media and 100 ppm arsenate sulfur media. By day 21, growth was observed in the following enrichments and each were transferred into fresh medium: EGno.1, BSno.2, VGno.1, SDno.1, BSno.1 and CSno.2 (all in Thermus 10 ppm arsenate media), as well as NNno.2 in sulfur media pH 2.5 0, 10, 100 and 1000 ppm

arsenate and BSno.3 in sulfur media pH 2.5 100 ppm arsenate. Transfers into fresh medium continued every couple of weeks there after. No growth was ever observed with samples plated onto semi-solid media.

Project title: Ecology of Hot Spring Microbial Communities

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Additional investigator(s): Mary Bateson, Thane Papke, Mike Ferris, Uli Nuebel

Objective: The general objective of our research is to understand the distribution and activity of microorganisms inhabiting microbial mat communities in geothermal effluents. At the moment, we are particularly interested in understanding the composition, structure and physiology of these mat communities, as models of microbial communities in general. We are using ribosomal RNA (rRNA), intervening transcribed spacer (ITS) and lipid biochemical cell components to identify community members. Our work relates to evolutionary microbiology in the sense that these gene sequences give phylogenetic information, and the association of lipids with their microbial sources helps us interpret the chemical fossil record produced by organic geochemists. In addition, we are attempting to evaluate whether the stable carbon isotope ratios of specific lipid biomarkers might help distinguish modern mat communities constructed by either cyanobacteria or green nonsulfur bacteria and hence their stromatolite counterparts in the fossil record.

Findings: During 2000 we made the following major observations: *Cyanobacteria, Synechococcus* populations in Mushroom Spring show a similar temperature distribution to those in Octopus Spring. We are currently examining their vertical distribution at four temperature-defined sites where we characterized light and chemical parameters using microsensors (Kühl). We succeeded in cultivating tens of *Synechococcus* isolates and are in the process of characterizing them genotypcially and phenotypcially with respect to possible adaptations to light and temperature. We are in the process of completing a study that suggests that genetically unique *Synechococcus* occur in hot springs around the world and even within Yellowstone. Green nonsulfur bacteria. We are completing a study of 16S rRNA-based diversity that suggests a remarkable diversity of such organisms in Mushroom Spring. We have continued experiments to evaluate the autotrophic metabolism of green nonsulfur bacteria by (1) using microsensors (Kühl) to demonstrate that the potential electron donors hydrogen and sulfide occur in the photic zone of mats containing cyanobacteria and green nonsulfur bacteria (i.e., Mushroom Spring and Tangerine Spring) in the morning and evening, and (2) conducting further 13C labeling studies at such times of day. We have extended our geochemical studies to investigate the 13C content of sugars and polyhydroxy alkanoic acids, which lead us to a competing hypothesis that heavy isotopic signatures of green nonsulfur-like bacteria in the mats may relate to the unique physiological situation involving major carbon flux through polysaccharides.

Project title: Isolation and Characterization of Thermophilic viruses from Yellowstone National Park

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Additional investigator(s): George Rice, Jamie Snyder, Blake Wiedenheft, Kenneth Stedman

Objective: To isolate and characterize thermophilic viruses from the thermal features of Yellowstone National Park.

Findings: Six different particle morphologies were found in *Sulfolobus* enrichment cultures grown at 80C and pH 3. Virus and virus-like particles were readily detected in a high proportion of enrichment cultures (43 percent), suggesting that viruses are a common feature of *Sulfolobus* species in YNP. Three of the particle morphologies are similar to viruses previously isolated from *Sulfolobus* species from Iceland and/or Japan. Three virus particle morphologies have not been previously observed from thermal environments. Some of these morphologies appear to be completely novel.

Despite the fact that the YNP SSV-like, SIRV-like, and SIFV-like particles have nearly identical morphologies to *Fusellovirus* (SSVs), *Rudiviruses* (SIRVs) and *Lipothrixviruses* (SIFV) isolated from Japan and Iceland, limited analysis of their genomic sequence indicates that they are only distantly related. This genomic diversity may reflect their long-term geographic isolation or it may be a function of adaptation to unique features of *Sulfolobus* species present in YNP. Ongoing studies are aimed at addressing the level of diversity within YNP as compared to related viruses present in other thermal regions of the world.

In contrast to the diversity of virus types, there does not appear to be a similar host diversity. The limited analysis of the 16S rDNA from several of the YNP enrichment cultures that were sequenced indicates that the Sulfulobus host species are nearly identical. The apparent contradiction of low host diversity with high virus diversity is intriguing. A thorough analysis of viruses present in *Sulfolobus* species is likely to provide unique insights into biochemical adaptations to life in extreme thermal environments. In addition, a detailed understanding of the viral replication cycle in *Sulfolobus* species will likely provide insight into cellular process present in Archaea and lead to a more thorough understanding of this unique domain of life.

ORNITHOLOGY

Project title: Movements and Survival of Bald Eagles Produced in YNP

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Additional investigator(s): Dr. George Montopoli, Peter Harmata

Objective: To determine long-term movements and survival of bald eagles banded as nestlings in YNP.

Findings: See (Harmata et al. 1999. Movements and survival of bald eagles banded in the Greater Yellowstone Ecosystem. *Journal of Wildlife Management* 63(3):781-793).

PALEONTOLOGY

Project title: Depositional Micro-environments and Preservation Potential of Plants and Arthropods in Recent and Fossil Hot-Spring Systems.

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Objective: Recent discoveries of exceptionally preserved early terrestrial plants and animals from the Early Devonian (400 Ma old) Rhynie chert hot spring complex (Aberdeenshire, Scotland, UK) has revealed the potential for exceptional fossilization (including soft tissues) within hot spring systems. The Rhynie complex consists of at least two separate vents, one of which exhibits geyserite splash texture. The current project extends the scope of the study to include recent silica-depositing systems within Yellowstone National Park as analogues for this fossil example. Collections of silica encrusted and silica entombed arthropods (primarily insects) will be examined alongside pre-existing water chemistry data and the geometry and topography of the hosting geothermal features to reveal processes involved in the entrapment and fossilization of organic debris.

Findings: Following on from our 1999 summer field season, our fieldwork continued to investigate the potential of different sub-environments in the geyser basins which promoted the preservation through silica encrusting and replacement of macro organic remains (plants and small arthropods). An interesting development in the research was the realization that threads of photosynthetic green alga in relatively cool pools (c. 30 degrees centigrade) could form sufficiently resilient frameworks which trapped and supported organic remains. Subsequently, silicification could take place on these framework templates.

Project title: A Preliminary Investigation of the Eocene Palynoflora of the Yancey Creek Drainage Basin, Yellowstone National Park, Wyoming

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Additional investigator(s): Melissa Stefos, Craig A. Chesner

Objective: To document Eocene palynoflora of Sepulcher formation. The purpose of this study is to examine the palynoflora of the Eocene Sepulcher Formation from a previously unstudied location near Yancey Creek, in the northern part of Yellowstone National Park. This is a preliminary investigation that hopes to document the presence of fossil pollen and spores at this site. The long range goal of this project includes extensive sampling resulting in more detailed floristic, paleoecological and stratigraphic analysis then is presently known.

Findings: Five rock samples for palynological analysis were obtained during July 2000 while the Eastern Illinois University Geology Field Camp was observing geological features in the park. The samples are dark brown or gray lithic rich sandstones, siltstones and shales. One gray sandstone specimen contains unidentifiable fossil plant fragments.

Some of the samples have been processed using a density separation and standard acetolysis chemical treatment with microscope slides prepared using a glycerine jelly medium. Thus far, most of the slides have proven to be barren of pollen. However, a few grains have been found and examination for statistically valid numbers of grains is continuing. Additional sample preparation is expected to lead to the discovery of enough palynomorphs to make preliminary scientific conclusions.

At this earliest stage of examination, a few grains of TCT *(Taxodiaceae-Cupressaceae-Taxaceae), Juglans,* and *Quercus* have been found. These findings are consistent with those of previous workers in other locations and fit well within the expected outcomes of the project. Details of this study will be posted on the Eastern Illinois University Geology Yellowstone National Park Research Program web site http://oldsci.eiu.edu/geology/camp/YNP/ynpres.htm.

Project title: Paleontological Survey of Yellowstone National Park

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Additional investigator(s): Vince Santucci

Objective: To develop a database of the fossil resources within Yellowstone National Park.

Findings: A significant amount of preliminary work has been accomplished. Vince Santucci has already published on the internet our current knowledge about the paleontology of Yellowstone National Park. The url is:http://www2.nature.nps.gov/grd/geology/paleo/yell_survey/index.htm. A manuscript on Eocene mammals from Yellowstone is in progress.

Range Management

Project title: Greybull Elementary Yellowstone Expedition: Rangeland Studies

Principal investigator: Mr. John Kunkel
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Objective: Observe changes in vegetative ground cover near the Specimen Ridge Trailhead.

Findings: Bare Ground, 22 percent; forbs, 35 percent; grasses, 27 percent; litter/scat, 23 percent; rock, 0 percent; shrubs, 0 percent. Location: N44 54.465' W110 19.508'.

RECREATION

Project title: The Evolving Social Construction of Wolves:
Exploring Yellowstone National Park Visitors' Social Interactions with *Canis lupus*

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Additional investigator(s): Michael Patterson, Jessica Montag

Objective: This study explores the visitor's perceptions of wolves, how these perceptions are shaped, the nature of the wolf watching experience and how these issues influence social conflict over wolf reintroduction and management. Using a social constructivist approach, this study is analyzing the in-depth interviews to explore 1) the public's social constructions of wolves in various contexts; 2) the nature of the recreational experiences individuals seek with respect to wolves; 3) how current social conflicts are affecting public perceptions; and 4) how new opportunities to interact with reintroduced wolf populations affect people's perceptions of wolves in the future. The goal of the study is to identify and describe the range of experiences and meanings associated with wolf watching.

Findings: From July 1999 through September 1999 21 in-depth interviews were completed. These interviews ranged from 10 minutes to 1 1/2 hours long, with the average interview lasting between 30-40 minutes. Another 10 interviews are expected to be completed between January and March of the year 2000. The interviews occur in the Lamar Valley, an area where wolf sightings are prevalent. Although analysis is ongoing, the nature of the insights that are being revealed include the dynamics of the wolf watching experience, the broader meanings of the Yellowstone wolves and how this experience is incorporated into the participant's life and into the total Yellowstone National Park experience.

VOLCANOLOGY

Project title: Monitoring Caldera Unrest at the Yellowstone Caldera:
A Global Positioning System (GPS) Crustal Deformation Study and Hot Springs Temperature Study by the Eastern Illinois University Geology Field Camp

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Additional investigator(s): William Toothill, Sid Halsor, Robert Jorstad

Objective: The purpose of our study is to monitor caldera unrest at the Yellowstone Caldera by annually collecting GPS data and hot springs temperatures from a network of data collection points. Although the data collected in this study can contribute to more detailed monitoring efforts of the caldera, the primary goal of this project is to provide an on-going, hands-on field experience for undergraduate geology majors at the Eastern Illinois University Geology Field Camp.

Findings: On 6/29-7/1/1999, and 6/28-6/29/2000 we deployed 3 Trimble 4000Ssi receivers at pre-established GPS control stations in the park. During a 48 hour interval each year, data was collected from 8 different control stations along a NW-SE traverse across the Yellowstone Caldera.

Following the 1999 survey, the data was processed and a set of 8 GPS baseline coordinates was established. During the 2000 GPS campaign, the 8 control stations were re-occupied. The data collected during the 2000 campaign was compared to the baseline data. Our results indicate that all of our data stations were displaced horizontally with components of southerly and westerly motion. Vertical motion, in the form of subsidence, also occurred at almost all stations. Southerly displacement of our stations was the least, ranging from 1 mm to nearly 2 cm, whereas westerly displacement ranged from about 1 cm to over 2 cm. With the exception of one control station, subsidence ranged between 3 and 10 cm. Maximum horizontal and vertical displacements occurred in the northern section of the network.

Although subsidence rates of up to 3 cm per year have been reported for the Yellowstone caldera, our subsidence values are significantly higher and may have associated errors. A review of the data collecting and processing procedures has not yet produced an insight into the potential source of error. A more detailed report of this study, including data tables and a map is available at:
http://wilkes1.wilkes.edu/~gis/yellowstone/yell2k/.

A second component of our study is monitoring hot springs temperatures at selected thermal areas within the Yellowstone Caldera. After consultation with park officials, three areas in the Lower Geyser Basin were chosen for our study including thermal areas along Rabbit Creek, White Creek, and Sentinel Meadows. On 6/28 and 6/29/2000, faculty members led small groups of students through these thermal areas to measure hot springs temperatures that will serve as baseline data for our study. Hot springs in the

Sentinel Meadows area mostly measured in the 90s, with several as high as 94 C. These consistently high temperatures qualified Sentinel Meadows as the overall hottest of our 3 study areas. Rabbit Creek and White Creek thermal features were more variable, with temperatures ranging from about 40 to 93 C. Maps and data tables for this study can be viewed at: http://oldsci.eiu.edu/geology/camp/YNP/ynpres.htm.

Project title: Eruption Observation of Selected Remote Geysers

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Additional investigator(s): Carlton Cross, Tara Cross

Objective: To obtain eruption interval and duration measurements of selected remote geysers at Shoshone Geyser Basin (Double Geyser, Frill Spring), Gibbon Geyser Basin (Phoenix Geyser, Oblique Geyser), Heart Lake Geyser Basin (Glade Geyser, Rustic Geyser), Lone Star Geyser Basin (Buried Geyser, Unnamed geyser SW of bridge).

Findings: Shoshone Geyser Basin: Double Geyser erupted every 4745-6196 seconds (mean of 34 intervals = 5319 sec); Frill Spring did not erupt between 07 July and 10 August 2000 and was very likely dormant during this time. Gibbon Geyser Basin: Phoenix Geyser erupted every 12874-14617 seconds (mean of 56 intervals = 14001 sec). Heart Lake Geyser Basin: Glade Geyser erupted every 95616-135605 seconds (mean of 8 intervals = 109683 sec). Lone Star Geyser Basin: Buried Geyser erupted roughly every 10-15 minutes; Unnamed geyser SW of the Lone Star-Shoshone Lake trail bridge was probably dormant between 12-14 August 2000.

Project title: Volcano Emissions

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Additional investigator(s): Dr. Terrence Gerlach, Mr. Michael Doukas, Mr. Richard Kessler

Objective: Survey and characterize carbon dioxide emissions from Yellowstone soils and thermal areas in order to identify possible areas of anomalous degassing from depth and to provide a baseline with which to compare future surveys of carbon dioxide in the event of volcanic unrest. The study involves airborne

measurements of carbon dioxide and other gases in the air above the park as well as ground measurements of carbon dioxide soil efflux within the park.

Findings: Several areas of carbon dioxide efflux have been measured within the park that are greater than what would be expected from normal biologic activity in the soil. In addition, several carbon dioxide plumes from various sources within the park were successfully measured in the air above the park in 1998 and 1999 utilizing sensitive instrumentation mounted in fixed-wing aircraft. Similar measurements utilizing helicopters as the airborne platform are anticipated. The detailed analysis of data is not yet complete.

Project title: Geochemistry, Biochemistry, and Stable Isotope Systematics of Sublacustrine Hydrothermal Vents in Yellowstone Lake

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Objective: The objectives of this study are to understand hydrothermal processes in sublacustrine hydrothermal vents in the context of the Yellowstone ecosystem and subaerial hydrothermal systems in and around Yellowstone National Park. In particular, we are using chemical composition, especially minor and trace elements and stable isotopes (H, C, N, O, and S), to understand processes of hydrothermal mineralization and to track potentially toxic and nutrient elements from hydrothermal vents into the micro- and macro-fauna of Yellowstone Lake and the greater Yellowstone ecosystem. To this end we have sampled lake waters, streams flowing into and out of Yellowstone Lake, sublacustrine sinter deposits and altered sediments, lake and cutthroat trout, bacterial mats and small crustaceans from vent localities, and similar materials from selected subaerial hydrothermal systems.

Findings: The geochemistry of Yellowstone Lake is strongly influenced by sublacustrine hydrothermal vent activity. The geothermal source fluid feeding the lake is identified by comparing the predicted Cl and hydrogen isotope data on water column, porewater, and vent samples from Yellowstone Lake. The chemical composition of sublacustrine hydrothermal vent fluids and the geothermal source fluid indicates strong enrichment of As, B, Cl, CO2, Ge, Hg, H2S, K, Li, Mo, Na, Rb, Sb, Si, and W.

The Cl concentrations indicate that Yellowstone Lake is about 1 percent geothermal source fluid and 99 percent inflowing stream water and that the flux is about 20 percent of the total geothermal water flux in Yellowstone National Park. With recent swath-sonar mapping studies that show numerous new hydrothermal features, Yellowstone Lake should now be considered one of the most significant geothermal basins in the park. Hg enrichments in hydrothermal vents and associated fauna contribute to elevated Hg concentrations in lake and cutthroat trout. Enriched Hg in cutthroat has potentially serious implications for grizzly bear, otter, eagle, and osprey populations that feed on cutthroat trout who spawn in the rivers.

Hydrothermal deposits occur on the lake bottom near active and presently inactive hydrothermal vents. Centimeter- to decimeter-sized siliceous deposits are cemented and recrystallized diatoms and represent pathways for hydrothermal fluid migration. A second major type of hydrothermal deposit comprises hard, porous siliceous spires that were discovered near Bridge Bay in 1997 by Dave Lovalvo and the UW Milwaukee group. At least 8-10 spires up to 7 m tall consist of diatom-rich areas and fibrous masses and globules of amorphous silica that could be of microbial in origin. Preliminary U-series dating gives an age of 11 ka for the silica spires. Bridge Bay spires formed in place by growth of chimney-like features from vigorous or long-lived lake-bottom hydrothermal vents. Chemical analyses indicate that siliceous vent deposits are almost always strongly enriched in As, Ba, Cs, Hg, Pb, and Sr, and are often enriched in Fe, Mo, Mn, Nb, Rb, Ta, Th, Tl, and W relative to normal Yellowstone Lake sediments. Oxygen isotope analyses of silica deposits indicate formation at temperature between 80 and 160 C. Chemical reaction modeling indicates that amorphous silica saturated fluid that vents into bottom waters at temperatures above 145 C can precipitate amorphous silica by mixing with cold, dilute lake waters.

Project title: Operation And Development of an Earthquake and Volcano Information System at Yellowstone and Ancillary Research on the Geodynamics of the Yellowstone Hotspot

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Objective: The primary objectives of the Yellowstone earthquake and volcano information system is to operate seismic and GPS networks, the Yellowstone seismic and GPS networks, necessary to monitor seismicity and ground deformation that may be related to volcanic, hydrothermal and tectonic earthquake activity. In addition ancillary research funded by the National Science Foundation titled "Geodynamics of the Yellowstone Hotspot" use the data and support in part the student and faculty research described herein.

Findings: In addition to routine network operations, notable efforts during the report period related to upgrading and maintaining seismograph and GPS stations against the harsh winter conditions of Yellowstone. Specific tasks included:

- Visiting 15 of the 22 stations of the Yellowstone seismograph network;
- Occupying five temporary digital seismographs at Lewis Lake, Nez Perce Creek, Canyon sewer plant, at the east entrance and at the NE entrance;
- Occupying of 60 temporary GPS receivers at sites established in the Yellowstone area;
- Moving the Little Thumb Creek seismic station out of the to be reclaimed gravel pit;

• Maintaining the continuous GPS stations at Lake, Mammoth, Old Faithful, Hayden Valley and White Lake;

• Continuing analysis on the systematic determination of local magnitudes;

• Submitting 19 years of Yellowstone seismic waveform data to the IRIS Data Management Center in SEED format;

• Submitting, several times per day, earthquake catalog data for the Yellowstone region to the Council of the National Seismic System's composite catalog;

• Assisting the National Park Service with long-term plans for implementing volcano and earthquake hazard assessment needs;

• Participating in the NPS-USGS workshop on Volcanoes in National Parks held at Mt. Lassen National Park;

• Discussing the development of a Yellowstone Volcano Observatory with the USGS Volcano Hazards group and NPS staff;

• Continuing analysis of ground deformation of the caldera using GPS and its relation to faulting and earthquakes of the nearby Hebgen Lake fault zone; and

• Presenting the findings and objectives of our research to Yellowstone NPS personnel, July 2000 as well as to several scientific meetings.

Project title: Water Temperature and pH Measurements

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Additional investigator(s): Dr. Steven Austin, Mr. Frank Sherwin

Objective: To take temperature and pH measurements in two separate hot pools in Yellowstone National Park and demonstrate high temperatures and varying levels of pH to an educational tour group.

Findings: We took temperature and pH measurements in Ojo Caliente runoff and in the road culvert near Roaring Mountain of Yellowstone National Park. Ojo Caliente runoff exhibited a higher temperature and opposite pH than Roaring Mountain.

WATER QUALITY

Project title: Yellowstone River Basin Study Unit National Water Quality Assessment

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Objective: The overall goals of the NAWQA Program are to 1) describe current water-quality conditions for a large part of the Nation's freshwater streams and aquifers; 2) describe how water quality is changing over time; and 3) improve our understanding of the primary natural and human factors affecting the water quality.

Findings: Samples were collected at several sites for bed sediment and fish tissue analysis. The samples were sent to the USGS National Water Quality Laboratory to be analyzed for trace elements and organics. A Water-Resources Investigations Report describing the results is in press at this time.

Project title: Reference Stream Monitoring: Long-term Trend Sites

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Objective: Collect long-term water quality, biological and stream habitat data at least-impacted streams in Yellowstone National Park. Reference stream data will be used as a benchmark for assessing water-quality, biological, and habitat conditions of test streams in the Middle Rockies ecoregion of Wyoming.

Findings: Monitoring of long-term reference sites is ongoing; no final report is available at this time. However, data from streams in YNP have been used to develop biological criteria for assessing aquatic life use attainment at other Middle Rockies ecoregion streams.

WILDLIFE MANAGEMENT

Project title: Winter Wolf Predation Rates and Prey Selection in an Elk-Bison System in Yellowstone National Park

Principal investigator: Dr. Robert Garrott
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Additional investigator(s): Rose Jaffe, Lee Eberhardt, Doug Smith, Kerry Murphy

Objective: The goals of this study were to quantify wolf predation rates and prey selection, and assess wolf predation impacts on the ungulate populations in the Madison, Firehole, and Gibbon drainages of Yellowstone National Park. Specific objectives included: 1) estimate time ungulates were subjected to wolf predation; 2) estimate winter ungulate abundance and composition; 3) estimate temporal patterns in predation, within and between winters; 4) describe prey selection patterns; and 5) estimate total ungulate offtake by wolves according to species, sex, and age class.

Findings: Data collection is complete and the thesis is being written for this study. Data were collected from November through May in the 1998-1999 and 1999-2000 winters. Triangulation and daily ground tracking of wolves was performed to collect data on wolf distribution and abundance, and locate kills. Necropsies were performed to ascertain species, sex, and age of kills to study wolf prey selection, and locations of kills were recorded to examine kill distribution. Ancillary data collected to investigate factors contributing to prey vulnerability included landscape attributes at kill, encounter, and failed attempt sites, and condition of prey. The amount of data collected was determined by daily wolf activity.

In the two years of study, wolves were monitored for a total of 345 days, radio signals were detected 218 days, and attempts to find wolf tracks off-road were made 178 days. Wolf tracks were followed and travel routes recorded 167 days for a total of 628 km. Areas of high use by wolves both years were Nez Perce Creek, Firehole Lake Drive, Twin Buttes, Sentinel Meadows, and Rabbit Creek.

The wolf preybase consisted of 300-900 bison and approximately 700 elk both winters. Eighty nine definite and 26 probable wolf kills were located during the study, including 59 elk calves, 31 cow elk, 10 bull elk, 1 unknown adult elk, and 15 bison: 13 calves, 1 cow and 1 unknown, totaling 115. Continuous non-denning season (mid-Novemberto mid-April) predation rates calculated for 98-99 and 99-00 winters showed an increase throughout both winters, with consistantly higher predation rates in the first winter compared to the second. Elk calves were the major prey item in both winters and prey switching was evident in both years. In the 1998-1999 winter, elk calves were taken at a steady increase November through February, decreasing in March. Adult elk and bison calves were taken at low levels all winter until increasing in March and April when elk calf kills began tapering. In the 1999-2000 winter, wolves began making kills in the study area later in the season than the previous year. Elk calves were taken consistently from

February through mid- March, decreasing in mid-April, while cow elk were taken increasingly throughout the winter. Bull elk and bison were negligible as prey items the second winter. Differences between winters included snow pack, above average in 1998-1999 verses below average in 1999-2000, and wolf pack size, 7 and 13 animals. Estimated prey offtake will be used to determine impacts of wolf predation on the ungulate population dynamics.

Project title: Winter Recreation Effects on Wildlife in Yellowstone National Park

Principal investigator: Dr. Robert Garrott
Contact: see above

Additional investigator(s): Dr. Scott Creel, Amanda Hardy

Objective: Our objective was to assess effects of winter recreation on wildlife populations in the Madison-Firehole-Gibbon drainages of Yellowstone National Park. We examined elk and bison distribution in relation to the winter road system, behavior, and stress hormone levels, along with winter season human activity types and levels. We compared distribution and behavior to similar data from a study conducted 20 years prior by K. Aune assessing winter recreation impacts on wildlife when winter visitation to YNP was significantly lower than current levels. Additionally, we compare behavior, distribution, and stress hormone levels to variation in human activity within the 1998-1999 and 1999-2000 winter season.

Findings: In the 1998-99 and 1999-2000 winter seasons, we collected data in the Madison-Gibbon-Firehole drainages of YNP. We repeatedly located 30-40 radio-collared cow elk and conducted bison surveys to obtain distribution and behavioral data relative to winter recreational activity. We collected fecal and snow-urine samples from radio-collared elk, unknown elk and unknown bison for analysis of glucocorticoid levels (a stress hormone indicator). We also conducted road, trail, and off-trail surveys documenting elk, bison, coyote, deer, moose, trumpeter swan and bald eagle sightings, along with group size, distance from road or trail, behavioral responses to human activity, and types of human activities present at each sighting. We have completed the lab work to derive fecal glucocorticoid levels and have compiled all field data for analysis. We are currently analyzing data and writing up our results. We have contributed one season of radio-collared cow elk stress hormone results and co-authored a paper for publication in Conservation Biology (paper pending acceptance as of February 2001). We plan to submit our completed findings to the NPS and for publication in Journal of Wildlife Management by the end of the summer (2001).

Project title: Coyote Density on Pronghorn and Bighorn Sheep Winter Range

Principal investigator: John Mack
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Additional investigator(s): McCrea Cobb

Objective: The Yellowstone pronghorn antelope herd is a historically and genetically significant group of animals that has served as a source for re-establishing pronghorn populations in other western states. During recent years, these antelope have experienced a precipitous decline in population. Several past studies support the conclusion that coyote predation can exert limiting influences on pronghorn populations. Little is currently known about the coyote populations inhabiting the Yellowstone pronghorns' critical winter range. The goal of this study is to estimate the density and numbers of coyotes and coyote packs in the Mammoth-Gardiner area using artificial howling surveys and scat transect surveys.

Findings: This study was approved and conducted during the winter and summer of 2000 and a similar study was conducted during the winter of 1998. During these studies, researchers identified and estimated the number of coyote packs in the study area. These past studies provide good data for comparison with the information gathered during continued surveys. We hope to repeat these studies using fundamentally identical methods this winter in order to build a database of information on coyote distribution and abundance in the study area.

Project title: Determining Bison Response to Mock Vaccination Approaches for Evaluating the Feasibility of a Remote Vaccination Program in Yellowstone National Park: Phase I

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Additional investigator(s): John Mack, Mark Biel, Travis Wyman, McCrea Cobb

Objective: We will conduct a series of mock vaccination approaches on Yellowstone bison during which we will measure the frequency and distance of flight, neutral reactions, or aggressive reactions of bison in response to these approaches. We will attempt to describe the relationship between bison behavioral reactions and field operational parameters (e.g. herd composition, number of personnel, mode of travel, distance to animals, weather, and location).

Findings: The primary product of Phase I is to determine the closest distance that park staff can safely approach bison on foot, horseback, snowshoe, vehicle, and snowmobile during each of the four sampling seasons while simultaneously minimizing stimulation of bison flight or aggressive responses. These data will then be incorporated into the development of optimum ballistic specifications (muzzle velocity, range, accuracy) for a remote vaccination gun. A final report will be generated by January, 2002.

Project Title: Epidemiology and Pathogenesis of Brucellosis in Yellowstone National Park Bison

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Objective: Determine the natural course of brucellosis in free-ranging bison. Determine modes of transmission. Provide information on the prevalence of infection and abortion.

Findings: We currently have 36 collared bison entering the sixth year of this project. Our focus will be on younger female bison coming into their first reproductive years, and on obtaining additional birth site information. Based on data we have collected to date bison apparently develop clinical brucellosis during their first pregnancy after exposure to the bacteria. Repeat reproductive failures, induced by brucellosis, appear to be uncommon. We have already begun removing radiocollars in 2000, and expect to remove more in February and spring months of 2001. The project will be completed and all radios will be removed in the fall of 2001.

Project title: Physiological Stress Responses, Aggression and Social Dominance in Yellowstone Wolves

Principal investigator: Dr. Scott Creel
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Additional investigator(s): Jennifer Sands

Objective: 1) Determine factors that affect stress hormone levels(glucocorticoids) in free-ranging wolves. This includes behavioral, ecological and anthropogenic influences; 2) Relate stress physiology to survival and reproduction; and 3) Relate stress physiology to social status.

Findings: We conducted two full winter seasons of field research in the winter of 1998-1999 and 1999-2000. We also continued field research this fall from November 2000 to February 2001. We have collected approximately 500 fecal samples since January 1999 from both known individuals, and where this was not possible, known pack. We have collected 500+ hours of observations of wolves that we are analyzing for

rates of aggressive interactions and dominance relationships. Our research has focused on the Druid, Rose Creek and Leopold packs in the Northern range of the park. We have finished running the glucocorticoid(GC) radioimmunoassays of the fecal samples collected and are beginning to analyze this data. We are currently conducting DNA analysis of fecal samples to determine genotypes and assign samples to specific individuals. We will also run radioimmunoassays for sex steroids (estrogen, estradiol and testosterone) beginning this spring. Our preliminary analysis indicates that higher ranking animals, in all three packs for both years studied, have higher GC levels than subordinates. Rank is a good indicator of GC levels, whereas rates of aggression are not. We will continue to examine other social, physiological, demographic and environmental variables and their association with GC levels.

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